201-12549

CHEMICAL MANUFACTURERS ASSOCIATION

COURTNEY M. PRICE
VICE PRESIDENT
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ORIGINAL

March 28, 2000

Carol Browner, Administrator
US EPA
PO Box 1473
Merrifield, VA 22116

Attn: Chemical Right-to-Know Program – Test Plan Submission from HERTG
Registration Number

Dear Ms. Browner:

The Chemical Manufacturers Association Petroleum Additives Panel (Panel) Health, Environmental, and Regulatory Task Group (HERTG) submits for review and public comment its test plan report, as well as related robust summaries, for the *"alkyl sulfide"* category of chemicals under the Environmental Protection Agency's High Production Volume (HPV) Chemical Challenge Program. The HERTG understands that there will be a 120-day review period for the test plan report and that all comments generated by or provided to EPA will be forwarded to the HERTG for consideration.

The alkyl sulfides in this category, which are used as petroleum lubricant additives, are characterized by having structural similarities and limited reactivity, low biological activity, and very low water solubility. Based upon the data reviewed in the attached report, the HERTG concludes that the physicochemical and toxicological properties of the proposed alkyl sulfide category members are similar and follow a regular pattern similar to the structural similarity. Thus, HERTG believes these five chemicals meet the EPA definition of a chemical category and will test them in accordance with the test plan summarized in the attached report. The five chemicals in the alkyl sulfide category are as follows:

- 2-propanol, 1-(tert-dodecylthio)- (CAS # 67124-09-8, referred to in this report as propanol/dodecylthio derivative)
- 1-decene, sulfurized (CAS # 72162-15-3, referred to in this report as decene derivative)
- 1-propene, 2-methyl-, sulfurized (CAS # 68511-50-2, referred to in this report as methyl propene derivative)
- Pentene, 2,4,4-trimethyl-, sulfurized (CAS # 68515-88-8, referred to in this report as trimethyl pentane derivative)
- Alkenes, C15-18 alpha-, sulfurized, (CAS # 67762-55-4, referred to in this report as C15-C18 alkene derivative).

MR34227

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Briefly, the HERTG's test plan for the alkyl sulfide category includes the following tests and computer modeling:

- Fugacity modeling – propanol/dodecylthio derivative (CAS # 67124-09-8); decene derivative (CAS # 72162-15-3) (mono- and disulfide variants); methyl propene derivative (CAS # 68511-50-2) (lengths y=3 and y=8); trimethyl pentane derivative (CAS # 68515-88-8) (homologues y=1 and y=4); and, C15-C18 alkene derivative (CAS # 67762-55-4 (mono- and disulfide variants)
- Biodegradability study – decene derivative (CAS # 72162-15-3)
- Photodegradation modeling – propanol/dodecylthio derivative (CAS # 67124-09-8), methylpropene derivative (CAS # 68511-50-2) and C15 – C18 alkene derivative (CAS # 67762-55-4)
- Acute fish toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8)
- Acute invertebrate toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8)
- Acute alga toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8)
- Reproductive/developmental toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8).

As HERTG developed this test plan, HERTG considered carefully and tried to limit how many animals might be required for tests included in the proposed plan and conditions to which the animals might be exposed. As noted above, a minimal amount of animal testing is proposed and, for those tests, HERTG believes the currently available scientific evidence suggests no significant toxicity will be demonstrated. As a result, HERTG believes that the concerns of some non-governmental organizations about animal welfare have been fully considered and that use of animals in this proposed test plan has been minimized.

HERTG previously committed to submit test plans for several other chemicals in 2000. Rather than submit all the reports at this time, HERTG wishes to wait until all comments are received on the alkyl sulfide report, as HERTG recognizes that comments submitted in response to this test plan report may impact HERTG's consideration of other test plan reports. HERTG anticipates that the test plan reports for its other chemicals committed to for 2000 will be submitted in May or June 2000.

Included in this package is a computer diskette that contains electronic copies of the HERTG's test plan report and accompanying robust summaries.

Thank you in advance for your attention to this matter. If you have any questions regarding the test plan report or the robust summaries, or HERTG's activities associated with the Challenge Program, please contact Kathleen Roberts, HERTG Manager. She can be reached at 703-741-5613 (telephone), 703-741-6091 (telefax) or Kathleen_Roberts@cmahq.com (e-mail).

Sincerely,

Courtney M. Price
Vice President
CHEMSTAR

cc: HERTG members

HIGH PRODUCTION VOLUME (HPV)

CHALLENGE PROGRAM

TEST PLAN

For

ALKYL SULFIDE CATEGORY

Prepared by
The Chemical Manufacturers Association
Petroleum Additives Panel
Health, Environmental, and Regulatory Task Group

March 28, 2000

LIST OF MEMBER COMPANIES IN THE HEALTH, ENVIRONMENTAL AND REGULATORY TASK GROUP

The Health, Environmental, and Regulatory Task Group (HERTG) of the Chemical Manufacturers Association Petroleum Additives Panel includes the following member companies:

Castrol Industrial North America

Chevron Chemical Company, LLC

CK Witco

Ethyl Corporation

ExxonMobil Chemical Company

Ferro Corporation

Infineum

The Lubrizol Corporation

Rhein Chemie Corporation

EXECUTIVE SUMMARY

The Chemical Manufacturers Association (CMA) Petroleum Additives Panel Health, Environmental, and Regulatory Task Group (HERTG), and its member companies, submit for review and public comment their test plan for the *"alkyl sulfide"* category of chemicals under the Environmental Protection Agency's High Production Volume (HPV) Chemical Challenge Program.

As discussed in the report that follows, these alkyl sulfides, which are used as petroleum lubricant additives, are characterized by having structural similarity and limited reactivity, low biological activity, and very low water volatility. Test data for members of the group show that they are of low concern for aquatic and mammalian toxicity, and, as a result, a reduced testing plan is scientifically justifiable to adequately characterize the category of chemicals.

Alkyl Sulfide Category. Relying on several factors specified in EPA's guidance document on "Development of Chemical Categories in the HPV Challenge Program," in which use of chemical categories is encouraged, the HERTG concluded that the following five closely related chemicals constitute a chemical category:

- 2-propanol, 1-(tert-dodecylthio)- (CAS # 67124-09-8, referred to in this report as propanol/dodecylthio derivative)
- 1-decene, sulfurized (CAS # 72162-15-3, referred to in this report as decene derivative)
- 1-propene, 2-methyl-, sulfurized (CAS # 68511-50-2, referred to in this report as methyl propene derivative)
- Pentene, 2,4,4-trimethyl-, sulfurized (CAS # 68515-88-8, referred to in this report as trimethyl pentane derivative)
- Alkenes, C15-18 alpha-, sulfurized, (CAS # 67762-55-4, referred to in this report as C15-C18 alkene derivative).

Structural Similarity. A key factor supporting treatment of these chemicals as a category is their structural similarity. All chemicals in this category consist of hydrocarbon chains (containing fully saturated bonds) with sulfide and polysulfide linkages. None of the chemicals within this category contain reactive (toxic) functional groups. Only one member of the category has a functional group – an alcohol group, which is not expected to be reactive.

Similarity of Physicochemical Properties. The similarity of the *physicochemical properties* of these substances parallels their structural similarity. All are dark colored viscous liquids intended for uses that require stability. The existing database for these substances shows they have limited reactivity, very low water solubility, and low vapor pressure. As a result, the members of the alkyl sulfide category have low potential for hydrolysis, extreme hydrophobicity, very low volatility, and (progressively at increasing molecular weights) limited ability to cross cell membranes. Consequently, they are expected to have low biological activity. What structural variability exists within the category is not expected to result in marked differences in

physicochemical properties, fate and transport characteristics, or in patterns of aquatic or mammalian toxicity. Available data and results of computer modeling support this assessment.

Fate and Transport Characteristics. Based on their physicochemical properties and molecular structures, the HERTG concluded that these chemicals are most likely to adsorb strongly to soil and sediments. To verify this conclusion, the HERTG will develop fugacity data on a number of homologues of the alkyl sulfide category chemicals. These chemicals are also expected to be resistant to hydrolysis and thus, to be stable in water. Compounds in the group are highly hydrophobic such that hydrolysis testing is not technically feasible and the lack of hydrolyzable moieties makes hydrolysis modeling unnecessary. Two of the five alkyl sulfides were subjected to biodegradability testing and found to be poorly biodegradable. The reason for this may be due to the high degree of branching in their alkyl chains. To determine whether there is potential for a higher degree of biodegradability with two of the members of this category that have linear alkyl groups, decene derivative (CAS # 72162-15-3) and C15-C18 alkene derivative (CAS # 67762-55-4), the HERTG will test the decene derivative (CAS # 72162-15-3), with test results to be bridged to two other compounds in the category. Results of this testing will be used to characterize the second material if shown to be significantly different than the existing data. Finally, while it is anticipated that alkyl sulfides will not absorb sufficient sunlight to photodegrade given their tendency to bond to soil, the HERTG plans to develop computer modeled data to adequately characterize the potential atmospheric oxidation potential of this category.

Toxicological Similarity. Review of existing published and unpublished test data for the alkyl sulfide category confirms the *similarity of aquatic and mammalian toxicity* among these five substances. In summary, based on available studies identified by the HERTG, these chemicals demonstrate low concern for aquatic and mammalian toxicity. These findings are expected based on the structure and physicochemical properties of these alkyl sulfide chemicals. In addition, the propanol/dodecylthio derivative (CAS # 67124-09-8) is expected to be the member of the group with the likely upper bound potential for toxicity.

> *Aquatic Toxicology.* Data on acute fish toxicity, acute invertebrate toxicity, and alga toxicity were reviewed. While the HERTG concluded that some additional aquatic toxicity testing is necessary as indicated in the test plan, the findings of available studies generally indicate low acute toxicity to fish and aquatic invertebrates, and low alga toxicity, when environmentally relevant test methods are used.
>
> *Mammalian Toxicology - Acute.* Data on acute mammalian toxicity (oral, dermal, and inhalation) were reviewed. Oral LD_{50} levels for all three substances tested were very high, indicating essentially no toxicity, even for the group member (compound in the group) most likely to show the upper bounds of toxicity. Similarly, acute dermal toxicity tests for three of the alkyl sulfide substances, including the compound most likely to show the upper bounds of toxicity, show essentially no toxicity. Inhalation toxicity test data were also reviewed for rats, mice, and guinea pigs. The results were again consistent showing low relative toxicity. The HERTG concluded that:

- the alkyl sulfide category has been generally well tested for acute mammalian effects;
- these tests show low acute toxicity;
- the studies include tests of the compound most likely to represent the upper bounds of acute toxicity; and
- no additional acute mammalian toxicity testing is necessary under the Challenge Program.

Mammalian Toxicology - Mutagenicity. Bacterial reverse mutation assay test data were available for four of the five members of the alkyl sulfide category. In each case the results were negative, both with and without metabolic activation. One of the five members of this category was tested in an *in vitro* chromosomal aberration assay. Again, all results were negative for clastogenicity, both with and without metabolic activation. *In vivo* chromosome aberration studies were available for two of the five alkyl sulfide substances, as well as a structurally similar analogue. All *in vivo* chromosome aberration data reviewed demonstrated that these alkyl sulfides are non-genotoxic, including the chemical in the group with the likely upper bound potential for genotoxicity. Thus, the HERTG concluded that:

- the alkyl sulfide category has been generally well tested for mutagenicity;
- these tests show low concern for mutagenicity;
- the studies include tests of the compound most likely to represent the upper bounds of mutagenicity; and
- no further mutagenicity testing is necessary under the Challenge Program.

Mammalian Toxicology - Subchronic Toxicity. The HERTG reviewed six repeated-dose studies with rats and/or rabbits for three of the five substances, including the substance with predicted upper bound potential for toxicity. No substance-specific toxicity was demonstrated. The changes that did occur in the laboratory animals were determined to be adaptive changes to liver or kidney effects that are not relevant to humans. Because of the consistency of results, the HERTG concluded that:

- the alkyl sulfide category has been generally well tested for repeated dose toxicity;
- these tests show low concern for repeated dose toxicity;
- the studies include a test of the compound most likely to represent the upper bounds of subchronic toxicity; and
- no additional repeated-dose toxicity studies are necessary under the Challenge Program.

Mammalian Toxicology - Reproductive and Developmental Toxicity. Although the alkyl sulfide category is well tested for other mammalian effects, the HERTG was unable to identify any published or unpublished reproductive/developmental studies considered adequate under the Program for compounds in the group. However, since the data on all other endpoints confirm the HERTG's expectation (based on structural similarity and similarity of physicochemical properties) that these chemicals are not biologically active, the HERTG believes no significant reproductive or developmental toxicity will be

demonstrated. Nevertheless, the HERTG plans reproductive and developmental toxicity testing for the propanol/dodecylthio derivative (CAS # 67124-09-8), the member of the group with the likely upper bound potential for toxicity so there will be reproductive and developmental toxicity data considered adequate under the Program for the substances of the group. If this test shows no developmental or reproductive toxicity, the HERTG believes the remaining members of the category would also show no developmental or reproductive toxicity. If this substance yields test results which are positive or equivocal, the HERTG will evaluate the need for additional reproductive testing in this category.

Conclusion. Based on the data reviewed in the report, the HERTG concludes that the physicochemical and toxicological properties of the proposed alkyl sulfide category members are similar and follow a regular pattern as a result of the structural similarity, and therefore meet the EPA definition of a chemical category. As a result, the HERTG believes these five chemicals constitute a category and will test them in accordance with the alkyl sulfide test plan summarized below.

Test Plan. The HERTG's test plan for the alkyl sulfide category includes the following tests and computer modeling:

- Fugacity modeling – propanol/dodecylthio derivative (CAS # 67124-09-8); decene derivative (CAS # 72162-15-3) (mono- and disulfide variants); methyl propene derivative (CAS # 68511-50-2) (lengths y=3 and y=8); trimethyl pentane derivative (CAS # 68515-88-8) (homologues y=1 and y=4); and, C15-C18 alkene derivative (CAS # 67762-55-4 (mono- and disulfide variants)
- Biodegradability study – decene derivative (CAS # 72162-15-3)
- Photodegradation study modeling – propanol/dodecylthio derivative (CAS # 67124-09-8), methylpropene derivative (CAS # 68511-50-2) and C15 – C18 alkene derivative (CAS # 67762-55-4)
- Acute fish toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8)
- Acute invertebrate toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8)
- Acute alga toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8)
- Reproductive/developmental toxicity study - propanol/dodecylthio derivative (CAS # 67124-09-8).

As HERTG developed this test plan, HERTG considered carefully how many animals might be required for tests included in the proposed plan and conditions to which the animals might be exposed. As noted above, a minimal amount of testing requiring use of animals is proposed and, for those tests, HERTG believes the currently available scientific evidence suggests no significant toxicity will be demonstrated. As a result, HERTG believes that the concerns of some non-governmental organizations about animal welfare have been fully considered and that use of animals in this proposed test plan has been minimized.

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1.0 INTRODUCTION

In March 1999, the Chemical Manufacturers Association (CMA) Petroleum Additives Panel Health, Environmental, and Regulatory Task Group (HERTG), and its participating member companies committed to address data needs for certain chemicals listed under the Environmental Protection Agency (EPA) High Production Volume (HPV) Chemical Challenge Program (Program). This test plan follows up on that commitment.

Specifically, this test plan sets forth how the HERTG intends to address testing information for the five substances listed in Table 1 and identified structurally in Figure 1. These five substances are propanol/dodecylthio derivative (CAS # 67124-09-8); decene derivative (CAS # 72162-15-3); methyl propene derivative (CAS # 68511-50-2); trimethyl pentane derivative (CAS # 68515-88-8); and C15-C18 alkene derivative (CAS # 67762-55-4).

As an integral part of its commitment to the HPV Challenge Program, HERTG has assembled and reviewed available data on these chemicals and determined that they constitute a "chemical category" as provided in the EPA guidance document entitled, "Development of Chemical Categories in the HPV Challenge Program." The following document provides the basis for that determination, indicates the findings of the data review process, and sets forth a proposed test plan to satisfy parts of the required test battery for endpoints without data that would be considered adequate under the program.

The basis for the HERTG determination that the five substances in this test plan should be treated as a chemical category (i.e., alkyl sulfides) under the HPV Challenge Program is set forth below.

EPA guidance on the HPV Challenge Program indicates that the primary purpose of the program is to encourage "the chemical industry . . . to voluntarily compile a Screening Information Data Set (SIDS) on all chemicals on the US HPV list." (EPA, "Development of Chemical Categories in the HPV Challenge Program," p. 1) At the same time, EPA recognizes that the "large number of chemicals to be tested [about 2800 HPV chemicals] makes it important to reduce the number of tests to be conducted, *where this is scientifically justifiable.*" (*Id.*, p. 1) [emphasis added] The next part of the guidance explains where this would be scientifically justifiable:

> One approach is to test closely related chemicals as a group, or category, rather than test them as individual chemicals. In the category approach, *not every chemical needs to be tested for every SIDS endpoint.* However, *the test data finally compiled* for the category must prove adequate to support a screening level hazard-assessment of the category and its members. That is, the *final data set* must allow one to estimate the hazard for the untested endpoints, *ideally* by interpolation between and among the category members. In certain cases, such as where toxicity is low and no upward trend is expected, extrapolation to the higher category members may be acceptable. (*Id.*, p. 1) [emphasis added].

EPA guidance goes on to state, "The use of categories is encouraged in the Challenge Program and will have a number of benefits." (*Id.*, p. 1) Among the benefits identified in the guidance for use of categories are that "a reduction in testing will result in fewer animals used to test a category of chemicals as opposed to doing each test on each individual chemical," and that "there will be . . . economic savings since less testing may be needed for chemicals considered as a category." (*Id.*, p. 1) That guidance also states that categories "accomplish the goal of the Challenge Program – to obtain screening level hazard information – through the strategic application of testing to the category." (*Id*, p. 2)

A similarly stated intent "to reduce the number of tests to be conducted, *where this is scientifically justifiable*" was articulated by the Agency in its draft guidance document titled, "The Use of Structure Activity Relationships (SAR) in the High Production Volume Chemicals Challenge Program." [emphasis added].

The EPA "Chemical Categories" guidance sets forth a definition of what constitutes a "chemical category, for the purposes of the Challenge Program". Specifically, that definition states that a chemical category under the HPV Challenge Program "is a group of chemicals whose physicochemical and toxicological properties *are likely to* be similar *or* follow a regular pattern as a result of structural similarity." (*Op. Cit*., p. 2) [emphasis added].

According to the guidance, what is important is that the "structural similarities [among members of the group] *may* create a predictable pattern *in any* or all of the following parameters: physicochemical properties, environmental fate and environmental effects, and human health effects." (Id., p. 2) [emphasis added]. Thus, it is not necessary for the chemicals in a category to be similar in all respects. Nor must there be conclusive proof that the chemicals in the postulated category will behave identically across all relevant parameters. All that is required for an acceptable category under the HPV program is that there be a *likelihood* of similarity of physicochemical and toxicological properties or a *likelihood* that the chemicals will in some pertinent respect follow a regular pattern as a result of their structural similarity.

In identifying the alkyl sulfide category, the HERTG followed the six-step process set out in the EPA guidance on category development. As the following information indicates, the alkyl sulfide category of chemicals put forth by the HERTG clearly satisfies the standards established in that guidance for use of a chemical category:

> Step 1: group structurally-similar chemicals into a putative category
>
> Step 2: gather relevant published and unpublished literature for each member of the category
>
> Step 3: evaluate the compiled data for adequacy in accordance with the EPA guidance documentation under the Program
>
> Step 4: construct matrices of SIDS endpoints versus category members arranged so as to indicate the structural progression of the category (in this case, by increasing molecular weight)

Step 5: evaluate the data to determine whether there is a correlation between category members for each SIDS endpoint.

Step 6: make available to EPA, and to the public for review, this test plan including the foregoing category definition and rationale and the following data assessment with the proposed testing scheme for the alkyl sulfide group of chemicals.

2.0 CHEMICAL DESCRIPTION OF ALKYL SULFIDE CATEGORY

The alkyl sulfide category consists of five closely related substances. The chemical names, CAS numbers, and structures for the members of the alkyl sulfide category are presented in Table 1 and Figure 1 (throughout the test plan the chemicals are arranged in the summary tables in order of increasing molecular weight). All five substances are derived from similar starting materials (i.e., alkanes/alkenes and sulfur), and all contain similar organic moieties linked by sulfur with linear, branching, or cyclic structures. Four substances include saturated long-chain hydrocarbons. Two of the substances contain mixtures of linear and cyclic alkyl sulfides. These substances can contain cyclic structures made up of sulfur and carbon, and the alkyl groups can be linear or branched. These structural similarities help explain the similarities in physicochemical properties, environmental fate, ecotoxicity, and mammalian toxicity and establish the justification of this group of materials as a category. Although propanol/dodecylthio derivative (CAS # 67124-09-8) contains a hydroxyl moiety, it is still a long-chain saturated hydrocarbon (a hexane and propyl chain) bridged by a sulfide with side chains consisting of six methyl groups. Consequently, it fits with the expected chemical and biological properties of this category.

The alkyl sulfides are dark colored viscous liquids. These substances range in molecular weight from 260 to 2,300 daltons with an average molecular weight of >500 daltons. As a group, the alkyl sulfides are extremely stable with very low water solubility and volatility. They are essentially unreactive because they are saturated and lack any available π (Pi) electrons that could interact with the nucelophilic center on biological molecules. All but one substance in this category lack functional groups that are potentially reactive or potentially hydrolyzable, such as alkyl halides, amides, thioamides, imines, carbamates, dithiocarbamates, carboxylic acid esters or other carbonyl functional groups, unsaturated carbon-carbon linkages (i.e., double or triple bonds between carbons), lactones, epoxides, urea groups, guanidine groups, nitro groups, nitrilo groups, azoxy groups, aziridines, azides, hydrazine groups, phosphate esters, heterocyclic functional groups, organic sulfate groups, or sulfonic acid esters. The substances in this category are extremely hydrophobic due to the long-chain aliphatic hydrocarbon structure and the absence of hydrophilic functional groups (as supported by the low estimated water solubility range for the members of this category; 0.5 mg/L to less than 0.001 mg/L). The low volatility of these materials is due in part to their high viscosity, low vapor pressure (10^{-3}-10^{-9} Pa at 20^{o}C) and range of molecular weights.

Due to the similar chemical characteristics of these five substances, they are expected to exhibit similar and predictable environmental fate and transport characteristics and patterns of mammalian and aquatic toxicity. The inherent stability of these structures suggests that they will

resist hydrolysis and photodegradation. The hydrophobic nature and low volatility of these substances suggests that they should partition to soil and sediment in the environment. The low water solubility of these substances suggests that exposure to aquatic organisms would be limited, which would decrease the likelihood of aquatic toxicity. The hydrophobic nature and molecular weights of these substances will decrease the likelihood of systemic toxicity to mammals. The low volatility of these substances limits the amount of these substances that can be inhaled, which will decrease the likelihood of respiratory toxicity. Furthermore, the chemical stability of these substances combined with the lack of an electrophilic moiety will decrease the likelihood of adverse effects that these substances may have upon the genetic material in cells.

3.0 EVALUATION OF AVAILABLE PUBLIC AND COMPANY DATA

3.1 ENVIRONMENTAL FATE DATA

3.1.1 Fugacity Modeling

Fugacity-based multimedia fate modeling compares the relative distribution of chemicals among environmental compartments (i.e., air, soil, sediment, suspended sediment, water, and biota). A widely used model for this approach is the EQC model[1].

There are multiple levels of the EQC model. In the document, "Determining the Adequacy of Existing Data", EPA states that it accepts Level I fugacity modeling to estimate transport/distribution values. In the same document the Agency states that Level III model data are considered "more realistic and useful for estimating a chemical's fate in the environment on a regional basis". The EQC Level I model utilizes input of basic chemical properties, including molecular weight, vapor pressure, and water solubility to calculate percent distribution within a standardized regional environment. EQC Level III uses these parameters to evaluate chemical distribution based on discharge rates into air, water, and soil, in addition to intermediate transport, advection, and degradation rates. EQC Level III will not be proposed for this evaluation because the physical properties of these chemicals do result in emissions or transport to air or water and appropriate emission levels are as yet unknown. Chemical distribution will be evaluated using a Level I model. Data developed using this level can then be used for simple comparative purposes across several chemical classes.

Some of the degradation rates for this class of chemicals will require an additional model to estimate physical/chemical properties from a structure. The model used for this purpose will be EPIWIN, version 3.02[2], which is also used by EPA and was developed by the Syracuse Research Corporation. EPIWIN includes algorithms for estimating all properties and rates needed for the application of the EQC model.

Nine basic chemical structures will be used for this evaluation and will represent the structures shown in Figure 1. All of the possible structure variations from these nine basic chemical structures will not be modeled, but for a number of chemicals the high and

[1] Equilibrium Criterion Model- Environmental Modeling Centre as developed by D. Mackay.
[2] Environmental Science Center- Syracuse Research Corporation- EPI for windows.

low molecular weight range will be evaluated in the models, as well as the mono- and di-sulfide linkages. Specifically, propanol/dodecylthio derivative (CAS # 67124-09-8) will be modeled as shown, and for the decene derivative (CAS # 72162-15-3), both the monosulfide and disulfide homologues will be evaluated. For the methyl propene derivative (CAS # 68511-50-2), dimethyl alkyl sulfide chain (noted as y in Figure 1) of lengths three and eight (i.e., y=3, y=8) will be used and for the trimethyl pentane derivative (CAS # 68515-88-8) methyl alkyl sulfide chain lengths of one and four will be modeled. Finally, for the C15-C18 alkene derivative (CAS # 67762-55-4), mono- and disulfide (straight chain) homologues with ethyl substituents (shown in parentheses in Figure 1) will be modeled.

All of these compounds have very low water solubility and low volatility. They also have very high log K_{ow} values and they bind strongly to organic carbon.

EQC modeling at Level I for the aforementioned chemicals will be conducted as part of the HPV test plan. Based on physical properties, it is expected that these chemicals are most likely to adsorb strongly to soil and sediments.

3.1.2 Hydrolysis

Hydrolysis is a reaction in which a water molecule (or hydroxide ion) substitutes for another atom or group of atoms present in an organic molecule. When an organic molecule undergoes hydrolysis, a nucleophile (water or hydroxide ion) attacks an electrophile and displaces a leaving group (e.g., halogen, phenoxide).[3] The lack of a suitable leaving group renders compounds resistant to hydrolysis. Examples of poor leaving groups are alcohols and sulfides. Potentially hydrolyzable groups include alkyl halides, amides, carbamates, carboxylic acid esters and lactones, epoxides, phosphate esters, and sulfonic acid esters (Neely, 1985[4]).

The five compounds in the alkyl sulfide category do not contain functional groups that are subject to hydrolytic reactions and have chemical components with a low potential for hydrolysis (Table 2).

OECD test guideline 111, *Hydrolysis as a Function of pH*[5], is used to assess the potential for a substance to hydrolyze in water. This test procedure cannot be applied to the HERTG alkyl sulfide category because of the low water solubility and analytical limitations discussed below. Aside from these limitations, determining the hydrolysis potential for these substances is not necessary because they do not contain organic functional groups that are susceptible to this physical degradative mechanism[6]. Therefore, these materials are expected to be stable in water.

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[3] W. Lyman et al. (1990) *Handbook of Chemical Estimation Methods.* Chapter 8.
[4] Neely, W.B. (1985) Hydrolysis. In: W.B. Neely and G.E. Blau. Eds. Environmental Exposure from Chemicals. Vol.I. CRC Press, Baca Raton, FL, USA. Pp.157-73
[5] Organization for Economic Cooperation and Development (OECD) (1993) OECD Guidelines for Testing of Chemicals. OECD. Paris, France.
[6] W.J. Lyman, W.F. Reehl, and D.H. Rosenblatt. (1982) Handbook of Chemical Property Estimation Methods. McGraw-Hill Book Co. New York, NY, USA.

For substances with water solubilities of less than $2x10^{-2}$ M, the OECD hydrolysis test procedure requires the preparation of a half saturatedaqueous solution of the test substance. For substances in the alkyl sulfide category, this would mean initial test solutions of 0.238 mg/L, based on the most water soluble member of this group of substances [EPIWIN[7], a computer program, was used to calculate the water solubility of the propanol/dodecylthio derivative (CAS # 67124-09-8)] to less than 0.0002 mg/L for the less water soluble members.

With regard to the test substance, the guideline states that the "analytical method must be sufficiently precise and sensitive to detect a reduction of 10 percent in the initial concentration." Initial aqueous concentrations of these substances, as required in the test guideline, are likely to be well below the level of analytical detectability. As a result, it would be unlikely that analytical methods would be sufficiently sensitive to detect a 10 percent reduction in test material concentration given the predicted low water solubility of the members in the alkyl sulfide category. As a result, even if these substances were susceptible to hydrolysis, this test could not be performed.

Based on the physicochemical characteristics of the chemicals in this category and the lack of organic functional groups, all the chemicals are expected to be stable in water. Therefore, no testing is necessary.

3.1.3 Biodegradability

Biodegradability is an important factor for determining the fate of chemicals in the environment because it provides a measure for the potential of compounds to be degraded by microorganisms. Chemical biodegradation involves a series of microbially-mediated reactions that may require many kinds of microorganisms acting together to degrade the parent chemical. There are several standard test methods available, each assesses potential biodegradability, based on a measured endpoint of which there are several. There are tests that only measure primary degradation (i.e., loss of parent chemical) or ultimate degradation (i.e., when the chemical is completely utilized resulting in the production of carbon dioxide, water, mineral salts, and microbial biomass). Primary degradation can be determined analytically by measuring dissolved organic carbon (DOC) for water-soluble chemicals by infrared absorbance, or by a chemical-specific method. Ultimate degradation (also called mineralization) is determined by measuring oxygen consumption or carbon dioxide evolution relative to the theoretical levels that can be achieved based on an elemental analysis of the chemical under investigation.

Tests have been developed for measuring biodegradability of chemicals under both aerobic and anaerobic (anoxic) conditions. However, currently only the aerobic tests

[7] EPIWIN. (1999) Estimation Program Interface for Windows, Version 3.02. Syracuse Research Corporation, Syracuse, NY, USA.

are used in classification and labeling of chemicals in the European Union, which allows for the use of data developed under OECD test guidelines, as well as other sources of standard guidelines.

As depicted in Table 3, two of the members of the alkyl sulfide category have been subject to biodegradability testing.

Propanol/dodecylthio derivative (CAS # 67124-09-8), the most water-soluble member of the category (0.475 mg/L), was subjected to conditions as specified in OECD guideline 301F, Manometric Respirometry Test. In the 28-day test, the measured oxygen demand was 5.9% of theoretical. Based on the test results, this compound exhibited a very slow rate of biodegradability.

Methyl propene derivative (CAS # 68511-50-2), potentially the second most soluble member ($\leq$0.000394 mg/L), was subjected to testing conditions as specified in OECD guideline 301B, Modified Sturm Test. In 28 days, 0.3% of the test material was converted to CO_2. Consequently, it was also assessed as exhibiting a very slow rate of biodegradability.

A biodegradability test will be conducted on the decene derivative (CAS # 72162-15-3) to determine if there is potential for a higher degree of biodegradability with two members of this category that have linear alkyl groups; i.e., decene derivative and C15-C18 alkene derivative (CAS # 67762-55-4). If the decene derivative shows significantly different results from the existing data, then the test results will be used to characterize these materials relative to their potential for a more rapid rate of biodegradability.

3.1.4 Photodegradation

Photodegradation is the degradation of a chemical compound as a result of absorption of solar radiation. Therefore, a prerequisite of photodegradation is that one or more bonds of the chemical compound in question has the ability to absorb ultraviolet (UV)/visible light in the 290 to 750 nm range. Light wavelengths longer than 750 nm do not contain sufficient energy to break chemical bonds, and wavelengths below 290 nm are shielded from the earth by the stratospheric ozone layer.

Four of the five substances in this category, including the decene derivative (CAS # 72162-15-3), methyl propene derivative (CAS # 68511-50-2), trimethyl pentane derivative (CAS # 68515-88-8), and C15-C18 alkene derivative (CAS # 67762-55-4), contain a polysulfide bond. These disulfide bonds are capable of absorbing light at a wavelength of 365 nm (i.e., within the wavelength range that may result in breakage of the disulfide bond). However, the strong bonding of these compounds to soil particles resulting from their hydrophobicity may reduce their tendency to absorb sufficient light energy to photodegrade.

The tendency of these alkyl sulfides to photodegrade will be evaluated by using the modeling program AOPWIN. This computer simulation of photo-oxidation is

recommended in the Agency's recently released structure activity review (SAR) guidance for HPV chemicals. Three of the five members of the category will be evaluated to estimate (1) rate constants for the atmospheric, gas phase reaction as mediated by photochemically produced hydroxyl radicals and (2) atmospheric half-lives based on hydroxyl radical concentrations. The members to be modeled are identified in Table 3. Methyl propene derivative (CAS# 68511-50-2) and C15-C18 alkene derivative (CAS# 67762-55-4) represent branched-chain and straight-chain compounds, respectively, that contain polysulfide bonds. Propanol/dodecylthio derivative (CAS #67124-09-8) contains a mono sulfide bond and it is the member with the lowest molecular weight and highest estimated water solubility. The remaining members contain a sulfide bond incapable of light adsorption at the requisite wavelength, so they do not need to be evaluated as they are expected to be non-photodegradable.

3.2. ECOTOXICOLOGY DATA

3.2.1 Aquatic Ecotoxicity Testing

3.2.1.1 Test Methodologies

Three test methodologies are commonly used to conduct aquatic toxicity tests; i.e., flow-through, static, and static renewal tests.

In *flow-through tests,* organisms are continually exposed to fresh chemical concentrations in each treatment level in the incoming water and there is greater assurance than with other test methods that the exposure levels and water quality remains constant throughout the test. Although flow-through testing is the preferred method, it is only applicable for chemicals that have adequate water solubility for testing. The alkyl sulfides cannot be tested by this method because they are all poorly soluble.

In *static tests,* organisms are exposed in still water that is not renewed. The chemical is added to the dilution water to produce the desired test concentrations. Test organisms are then placed in the test chambers and there is no change of water at any time during the test. There is less assurance that the test concentrations test organisms are exposed to will remain constant because test material can be adsorbed onto test chambers, degraded, volatilized, or otherwise changed during the test. Nevertheless, due to limitations of other test systems for non-volatile materials, the static test has been widely used by the scientific community.

The *static-renewal test* is similar to a static test because it is conducted in still water, but the test solutions and control water are renewed periodically, usually every 24 hours. Daily test solution renewal ensures that the exposure concentrations are more likely to be stable throughout the test. This is the preferred method for conducting aquatic toxicity tests for compounds such as the alkyl sulfides on fish. Daily renewals cannot be done in the algae test, and usually not in *Daphnia* tests, because the process of separation and replenishment

would cause a discontinuity in the alga growth rate and it can stress, coat, or entrap *Daphnia* in any surface film during renewals. OECD considers static and static renewal tests appropriate for testing poorly soluble chemicals like the alkyl sulfides provided that test solution preparation use water accommodated fraction or water soluble fraction methods.

3.2.1.2 Test Solution Preparation

Alkyl sulfides are poorly water–soluble substances and it is not possible to prepare exposure solutions for aquatic toxicity testing by direct addition of measured quantities of test material to water. Two methods are used to prepare solutions of poorly soluble materials for aquatic toxicity testing:

- *Water accommodated fraction* (WAF) – This is a method in which the test solution contains only that fraction of the test material (organic phase) which is retained in the aqueous phase after a period of stirring long enough to reach equilibrium, followed by a sufficient time for phase separation. The WAF (aqueous phase) will contain soluble components of the test material at levels that will be dependent on the test material loading (the amount of material added to the aqueous medium). The resulting WAF is used in the aquatic toxicity test. Ideally, a WAF consists of a water-soluble extract of test material, but it can also include a stable micro-emulsion or contain small amounts of suspended matter.

- *Water soluble fraction* (WSF) – This is a method in which a WAF is either filtered, centrifuged, or allowed to settle for a greater length of time (24 hours) than with the WAF method to remove suspended matter from the aqueous phase before being used in the aquatic toxicity test.

3.2.1.3 Reporting Toxicity Results

In both WAF and WSF tests, test material concentrations are expressed as loading rates (i.e., defined as the weight of test material added per unit volume of test medium)[8]. For fish tests, endpoints can be expressed as median lethal loading rate (LL_{50}) when lethal effects occur to 50% of the test population or in cases where no lethal effects are observed at all loadings tested, LL_0. In both cases, results can be expressed in mg/L and in studies where no effects are observed, the result is expressed as LL_0 = the highest loading tested. For invertebrate and alga tests, endpoints are expressed as median effective loading rate (EL_{50}) or EL_0 in mg/L as discussed above.

Loading rates allow poorly water-soluble complex substances such as the alkyl sulfides to be compared to more readily soluble substances and /or pure chemicals on an equal basis (OECD[8]). To allow comparison, the toxicity value is expressed as the amount of test material added per unit volume of water.

[8] Organization for Economic Cooperation and Development (OECD) (1999) Draft Guidance document on Aquatic Toxicity Testing of Difficult Substances. OECD, France.

If test material exposure levels are analytically measured in the test, the endpoints can also be expressed as median lethal concentration (LC_{50}) or median effective concentration (EC_{50}) in mg/L. EC/LC_{50}s are often not reported because it is very difficult to accurately measure test material exposure levels that can be below 1.0 mg/L.

NOTE: Test results are expressed as EC_{50} or LC_{50} even though the tests used WAF or WSF methodology and based on current reporting procedures would be reported as loading rates. In the interest of maintaining consistency between this document and the test reports, the toxicity results have been presented as they were originally reported and converted to lethal loading rate values as described above, which correctly represent the test procedures.

3.2.2 Aquatic Toxicity of the Alkyl Sulfide Category

In general, the toxicity of a substance is limited by absorption into the organism and movement to the target organ(s). Characteristics such as smaller molecule size and a lesser degree of ionization increase the ability of the substance to cross biological membranes. Furthermore, the soluble quantity of a compound in water represents the fraction responsible for toxicity; aquatic toxicity is therefore limited by the water solubility of a compound. For the members of the alkyl sulfide category, the lack of toxicity is due to their low water solubility. Therefore, the substances in the group have limited bioavailability to aquatic organisms. While all group members have low water solubility, relative water solubility is directly linked to alkyl chain length. For equal chain lengths, branched chains are presumed more water soluble than straight chains. Therefore, based on chain length and branching, the lowest molecular weight member of the category, the propanol/dodecylthio derivative (CAS # 67124-09-8) is predicted to be the most water-soluble member of the category and the chemical most likely to represent the upper bounds of aquatic toxicity.

Table 4 summarizes acceptable aquatic toxicity studies and proposed information for the alkyl sulfide category under the HPV Program.

3.2.2.1 Fish Acute Toxicity

The fish acute toxicity test establishes the lethality of a substance to a fish after a 96-hour exposure period. Tests on a member of the alkyl sulfide category assessed in this evaluation were performed in accordance with OECD guideline #203, *Fish, Acute Toxicity Test*.

Because propanol/dodecylthio derivative represents the lower range of molecular size for this category, this material may demonstrate acute toxicity for this endpoint. Therefore, the acute fish toxicity of propanol/dodecylthio derivative (CAS # 67124-09-8) will be determined.

Methyl propene derivative (CAS # 68511-50-2) was tested using WAF and WSF methodology. The results show that this product will not demonstrate acute

toxicity to fish at loading rates of 1,000 mg/L and 10,000 mg/L, as demonstrated using WAF and WSF methodology, respectively. This most likely is due to its high molecular weight and correspondingly low water solubility. These are also characteristics of the three remaining members of this category. Therefore, the acute toxicity data developed for methyl propene derivative (CAS # 68511-50-2) will be used to assess the toxicity of decene derivative (CAS # 72162-15-3); trimethyl pentane derivative (CAS # 68515-88-8); and C15-C18 alkene derivative (CAS # 67762-55-4) to fish.

3.2.2.2 Invertebrate Acute Toxicity

The acute invertebrate test establishes the lethality of a substance to an invertebrate, typically a daphnid (*Daphnia magna*), after a 48-hour exposure period. The tests included in this evaluation (Table 4) were performed in accordance with OECD guideline #202, *Daphnia sp*., Acute Immobilization Test and Reproduction Test. Acute invertebrate toxicity was tested using WAF methodology.

Because propanol/dodecylthio derivative (CAS # 67124-09-8) represents the lower range of molecular size for this category, this material may demonstrate toxicity for this endpoint. Therefore, the acute invertebrate toxicity of propanol/dodecylthio derivative (CAS # 67124-09-8) will be tested.

In comparison, the remaining members of this category are expected to demonstrate toxicity equivalent to methyl propene derivative (CAS # 68511-50-2) because of their similar higher molecular weights. Existing data for methyl propene derivative (CAS # 68511-50-2) show that this material will not demonstrate acute toxicity to *Daphnia magna* at a loading rate of 1,000 mg/L using the WAF methodology. This is likely due to its high molecular weight and correspondingly low water solubility. Therefore, the acute toxicity data developed for methyl propene derivative (CAS # 68511-50-2) will be used to assess the toxicity of decene derivative (CAS # 72162-15-3); trimethyl pentane derivative (CAS # 68515-88-8); and C15-C18 alkene derivative (CAS # 67762-55-4) to invertebrates.

3.2.2.3 Alga Toxicity

The alga growth inhibition test establishes the potential of a substance to inhibit alga growth, typically using the freshwater unicellular green algae, *Pseudokirchneriella subcapitata* (formerly called *Selenastrum capricornutum*), after a 96-hour exposure period. The test included in this evaluation (Table 4) was performed in accordance with OECD guideline #201, *Alga, Growth Inhibition Test*. Alga growth inhibition was evaluated using WAF methodology. The results are depicted in Table 4.

No data are available for propanol/dodecylthio derivative (CAS # 67124-09-8). Because propanol/dodecylthio derivative (CAS # 67124-09-8) represents the lower range of molecular size for this category, it is anticipated that this material

is the most likely to demonstrate toxicity. Therefore, it is proposed that the acute alga toxicity of propanol/dodecylthio derivative (CAS # 67124-09-8) be determined.

Methyl propene derivative (CAS # 68511-50-2) was tested using WAF methodology. Existing data for methyl propene derivative (CAS # 68511-50-2) show this chemical demonstrates toxicity to alga at loading rates greater than 100 mg/L using WAF methodology. The remaining members of this category are expected to demonstrate toxicity equivalent to methyl propene derivative (CAS # 68511-50-2) because of their similar higher molecular weights and correspondingly low water solubility. Therefore, the toxicity data developed for methyl propene derivative (CAS # 68511-50-2) will be used to assess the toxicity of decene derivative (CAS # 72162-15-3); trimethyl pentane derivative (CAS # 68515-88-8); and C15-C18 alkene derivative (CAS # 67762-55-4) to alga.

3.3 MAMMALIAN TOXICOLOGY DATA

3.3.1 Physicochemical Properties Relevant to Mammalian Toxicity

Typically, for a xenobiotic[9] to be biologically active it must be able to:

- cross biological membranes in order to reach target tissue/cells in an organism (e.g., either by passive diffusion or active transport via carrier proteins). According to basic principles of pharmacology and toxicology, lipophilicity generally enhances the ability of chemicals to cross biological membranes (i.e., lipophilic compounds are more readily absorbed across biological membranes than hydrophilic or charged chemical moieties). However, for large lipophilic compounds, molecular size becomes a critical limiting determinant (i.e., small lipophilic compounds more readily traverse biological membranes than do large lipophilic materials);
- be transported within the systemic circulation to target cells, provided the target cells are not located at the initial site of entry or contact. Contrary to absorption, which is favored by lipophilicity, basic tenets of pharmacology and toxicology maintain that systemic distribution or transport within the body is enhanced or facilitated by hydrophilicity (i.e., a compound that is hydrophilic may be more readily transported via plasma). At the same time, hydrophilicity enhances urinary excretion and decreases the biological half-life of a xenobiotic within an organism
- be biologically active or be activated by enzyme systems, such as mixed function oxidase (cytochrome p450 or p448 systems). Biotransformation of a xenobiotic to increase its water solubility for the purpose of enhancing excretion may generate reactive intermediates.
- interact chemically or physically with target cells or receptors (e.g., the chemical may interact with a biological receptor molecule or react with biological macromolecules and the interaction or reaction, such as oxidation or epoxidation, may result in disruption of normal biological function at that target site). In acute or repeated-dose toxicity studies this interaction may occur because the substance or a metabolite

[9] A xenobiotic is any chemical compound that is foreign to a living organism.

mimics an endogenous substrate producing an adverse effect. In mutagenicity studies, this interaction may cause the impediment or alteration of DNA replication or a chemical reaction with DNA base pairs.

The similarity in physicochemical properties of the five alkyl sulfides which comprise this category suggest that the mammalian toxicity of all members of the category will be similar. Further, the structural features and properties of the members of the category suggest that the alkyl sulfides will be unable to reach target organs or, if they do, that they are not likely to produce toxic effects. The hydrophobic nature of these substances may favor penetration of biological membranes, but their subsequent transport via the circulatory system to target cell or receptors will be limited due to these same hydrophobic properties and the general absence of functional groups and reactive sites where biotransformation reactions might enhance hydrophilicity. The low volatility of these substances suggests that very low amounts of these materials will be available for absorption via inhalation. The high viscosity of these substances suggests that it will be difficult to generate high concentration of respirable particles in the air. Four of these compounds [i.e., methyl propene derivative (CAS # 68511-50-2), trimethyl pentane derivative (CAS # 68515-88-8), decene derivative (CAS # 67762-55-4), and C15-C18 alkene derivative (CAS # 72162-15-3)] consist of saturated carbon-carbon linkages and extremely stable polysulfide bonds; consequently, biological activation via epoxidation, O-insertion, or hydrolysis is thermodynamically unfavorable and is unlikely. In addition, these four substances lack functional groups that are potentially reactive or susceptible to hydrolysis as stated in Section 3.1.2.

The member of the category with the lowest molecular weight (propanol/dodecylthio derivative; CAS # 67124-09-8) contains a hydroxyl group.[10,11] The hydroxyl group could be chemically reactive under some circumstances, but the level of delocalized electrons within the molecule stabilizes it. The saturated linkages limit the possibility of a strong sigma negative charge on the hydroxyl group (which is a weak electrophile). Furthermore, the hydroxyl group confers a greater degree of hydrophilicity. It is important to note that while the hydroxyl group provides a potential site of oxidation to form a carbonyl or carboxylic acid moiety, the hydroxyl group is also a target for glucuronidation and sulfation reactions. These types of biotransformation reactions are generally considered detoxification pathways that lead to inactivation of a potentially reactive site and enhance urinary excretion. Consequently, this substance is expected to share the same toxicological properties as the rest of the category. At the same time, these features (i.e., smallest member of the category, presence of the single, potentially reactive functional group within the category, and expected greatest degree of relative bioavailability within the category) are such that if any member of the category may show mammalian toxicity, it is more likely to be the propanol/dodecylthio derivative than any other in the category. Thus, it serves as a "marker chemical" that establishes the potential upper bound potential for mammalian toxicity for the entire group. Assuming

[10] "The Use of Structure-Activity Relationships (SAR) in the High Production Volume Chemicals Challenge" Program (1999) EPA Chemical Right-to-Know website August 26, 1999.

[11] Federal Drug Administration, Toxicological Principles for the Safety Assessment of Direct Food Additives, Appendix 1.

the mammalian toxicity of this substance is similar to that of the remaining four, then the designation of these five alkyl sulfides as a category, as defined by EPA, is scientifically justified.[12]

3.3.2 Acute Mammalian Toxicity of the Alkyl Sulfide Category

3.3.2.1 Acute Toxicity Test Methodology

Acute toxicity studies investigate the effect(s) of a single exposure to a relatively high dose of a substance. Potential routes of exposure for acute toxicity assays include oral, dermal, and inhalation. Oral toxicity assays are conducted by administering test material to fasted animals (typically rats or mice) in a single gavage dose. Acute dermal toxicity tests are conducted by administering test material to the shaved skin on the back of the test animal (typically rats or rabbits) and allowing the test material to stay in contact with the skin application site for a specific duration (usually 24 hours). Acute inhalation toxicity assays are conducted by exposing test animals (typically rats) in a controlled atmosphere to a fixed air concentration of the test substance for a specific duration (typically 4 hours). The test material is either generated as a vapor or intentionally aerosolized into respirable particles, then metered into the exposure air at the desired concentration. Preferably, inhalation toxicity studies are conducted using either nose-only or head-only exposure to minimize potential confounding effects resulting from whole-body exposure. This methodology may lead to over-prediction of inhalation toxicity hazard (e.g., increasing the body-burden of the test material through skin absorption or ingestion (oral exposure) of test material as a consequence of grooming both during and after the intended exposure period).

Historically, lethality is a primary end-point of concern in acute toxicity studies and the traditional index of acute toxicity potency is the medial lethal dose that causes mortality in 50 percent of the test animals (LD_{50}). In acute inhalation studies, the traditional metric of potency is the median lethal concentration of the test material in air that causes mortality in 50 percent of the test animals(LC_{50}). In addition to lethality, acute toxicity studies also provide insights regarding potential systemic toxicity through careful observation and recording of clinical signs and symptoms of toxicity as well as through detailed examination of tissues and organ systems.

Typically, acute oral and dermal toxicity studies are conducted using a limit dose of 5000 and 2000 mg/kg body weight, respectively, and acute inhalation toxicity studies are conducted using a limit dose of 5 mg/L for 4 hours (according to OECD and EPA testing guidelines). Recently, harmonized EPA testing guidelines (August 1998) have set the limit dose for both oral and dermal acute toxicity studies at 2000 mg/kg body weight, while the recommended limit concentration for acute inhalation studies has been set at 2mg/L for 4 hours. The

[12]EPA guidance document: "Development of Chemical Categories in the HPV Challenge Program" (Last Revision, August 25, 1999).

limit dose test method minimizes the number of animals tested by exposing a single group of animals to a large dose (the limit dose) of the test substance. If less than 50 percent mortality is observed at the limit dose, no further testing is needed. A test substance that shows effects at a concentration greater than the limit dose is considered essentially nontoxic. If compound-related mortality is observed, then further testing may be necessary.

3.3.2.2 Summary of Available Data

Acute toxicity information considered adequate under the Program for the alkyl sulfide category are summarized in Table 5.

3.3.2.2.1 Acute Oral Toxicity

Acute oral toxicity studies in rats considered adequate under the Program are available on three of the five chemicals in the alkyl sulfide category: propanol/dodecylthio derivative (CAS # 67124-09-8), methyl propene derivative (CAS # 68511-50-2), and trimethyl pentane derivative (CAS # 68515-88-8). The acute oral toxicity tests for these chemicals were performed in accordance with OECD Guideline 401. The oral LD_{50} for all three substances was greater than the 5,000 mg/kg limit dose indicating that these substances have relatively low toxicity.

Summary: All three compounds referenced above were essentially nontoxic upon acute oral exposure (i.e., LD_{50} greater than the limit dose of 5000 mg/kg body weight), including the marker compound, propanol/dodecylthio derivative (CAS # 67124-09-8). In addition, the consistency of information on acute oral toxicity further supports treatment of all five alkyl sulfides as a chemical category within the HPV Program.

3.3.2.2.2 Acute Dermal Toxicity

Three of the substances in the alkyl sulfide category were adequately tested for use under the Program for acute dermal toxicity in rabbits: propanol/dodecylthio derivative (CAS # 67124-09-8), trimethyl pentane derivative (CAS # 68515-88-8), and C15-C18 alkene derivative (CAS # 67762-55-4). These studies were conducted in accordance with OECD Guideline 402. No mortality or treatment-related clinical signs of toxicity or gross lesions were observed for any substance when tested at the limit dose of 2000 mg/kg. Thus the dermal LD_{50} was greater than 2000 mg/kg for all three substances, including the designated marker chemical within the group, the propanol/dodecylthio derivative (CAS # 67124-09-8).

Summary: Similar to the acute oral toxicity results, all three alkyl sulfides compounds referenced above were essentially nontoxic following acute dermal exposure (i.e., LD_{50} greater than the limit dose of 2000 mg/kg body weight), including the marker compound, propanol/dodecylthio derivative (CAS # 67124-09-8). As with the acute oral toxicity results, the consistency of

data on acute dermal toxicity further substantiates treatment of all five alkyl sulfides as an HPV chemical category.

3.3.2.2.3 Acute Inhalation Toxicity

Two of the substances in the alkyl sulfide category have been adequately tested under the Program for acute inhalation toxicity: methyl propene derivative (CAS # 68511-50-2) and trimethyl pentane derivative (CAS # 68515-88-8). These tests were conducted according to OECD Guideline 403. The methyl propene derivative (CAS # 68511-50-2) was tested in rats, while the trimethyl pentane derivative (CAS # 68515-88-8) was tested in rats, mice, and guinea pigs.

In a study conducted with the methyl propene derivative (CAS # 68511-50-2), rats were exposed to a vapor of the test material at three concentrations. The highest dose tested was 0.39 mg/L with a whole-body exposure for 4 hours. No mortality was noted, and all animals fully recovered following depuration. No significant clinical signs were noted after the initial post-exposure observations at any dose level. No treatment-related macroscopic or microscopic findings were noted. The LC_{50} for the methyl propene derivative (CAS # 68511-50-2) was greater than 0.39 mg/L (highest dose tested).

Acute inhalation toxicity testing of the trimethyl pentane derivative (CAS # 68515-88-8), involved exposure to an aerosol of the test material. In one study, the LC_{50} was >5.6 mg/L for male rats and equal to 2.17 mg/L for female rats. No treatment-related gross lesions were noted in surviving rats.

Two other acute inhalation studies with aerosolized trimethyl pentane derivative (CAS # 68515-88-8) were conducted in rats, mice, and guinea pigs (rats, mice, and guinea pigs in one study and guinea pigs and mice in the second study). One study noted mortality in 3 out of 10 rats, 1 out of 10 mice, and 1 out of 10 guinea pigs at the maximum attainable concentration of 4.3 mg/L. No treatment-related clinical signs were noted for mice and guinea pigs. In another study in which mice and guinea pigs were also tested at the maximum attainable concentration of 4.3 mg/L, no mortality and no treatment-related clinical signs or gross lesions were noted. For mice and guinea pigs, the LC_{50} is considered to be >4.3 mg/L (maximum attainable concentration). The LC_{50} for male rats in this study is considered to be >4.3 mg/L, and the LC_{50} for females is <4.3 mg/L.

Summary: For the two compounds referenced above for acute inhalation toxicity, both exhibited relatively low toxicity (LC_{50}s were all above the maximum achievable concentration). The relatively low maximum achievable air concentrations are consistent with the low vapor pressures and the physical limitations of aerosolizing high viscosity materials. The administered dose via inhalation is very low compared to other routes of exposure (e.g., oral or dermal), thus decreasing the potential exposure of these

compounds via inhalation pathways. Inhalation is not considered a relevant route of exposure for the alkyl sulfides because of the limited volatilization and aerosolization potential of these compounds.

3.3.2.3 Data Assessment and Test Plan for Acute Mammalian Toxicity

In total, eleven acute toxicity studies considered adequate under the HPV Program have been conducted upon the alkyl sulfide chemical category. These studies have involved four different animal species; oral, dermal, and inhalation routes of exposure; and four of the five members of the category. The data consistently demonstrate low or no acute toxicity following acute oral, dermal or inhalation exposure. The HERTG conclude that the acute toxicity findings support the expectation, based on structural, physicochemical, and toxicological similarities, that these 5 compounds should be treated as a category. The HPV Challenge Program requires that either an acute oral, dermal, or inhalation test (preferably oral) be performed or bridged to each member chemical of a category. Although acute toxicity testing sufficient for the Program has not been conducted on the decene derivative (CAS # 72162-15-3), the structural features and physicochemical properties of the derivative are sufficiently similar to those of the other members of this category. Thus the decene derivative (CAS # 72162-15-3) is expected to exhibit acute toxicity findings similar to those of the other alkyl sulfides in this category, and, therefore, additional testing of the decene derivative (CAS # 72162-15-3) is not needed. The HERTG has concluded that the acute toxicity of the category has been evaluated adequately with respect to all acute toxicity HPV endpoints. No additional acute toxicity testing is proposed for purposes of the HPV Challenge Program.

3.3.3 Mutagenicity of the Alkyl Sulfide Category

3.3.3.1 Mutagenicity Test Methodology

Genetic toxicology is concerned with the effects of substances on genetic material (i.e., DNA and chromosomes). Within genetic material, the gene is the simplest functional unit whose essence is composed of DNA. Mutations are generally nonlethal, heritable changes to genes which may arise spontaneously or as a consequence of xenobiotic exposure. Genetic mutations are commonly measured in bacterial and mammalian cells. The simplest test systems measure the occurrence of a base-pair substitution mutation in which a single nucleotide is changed followed by a subsequent change in the complementary nucleotide on the other DNA strand. Frameshift mutations occur following the deletion or insertion of one or more nucleotides, which then changes the "reading frame" for the remainder of the gene or multiple genes. Genetic testing for these types of point mutations is generally accomplished by *in vitro* cellular assays for forward or reverse mutations. A forward mutation occurs when there is a detectable change in native DNA whereas a reverse mutation occurs when a mutated cell is returned to its initial phenotype. Both base-pair substitutions and frameshift mutations are routinely measured in bacterial cells by measuring the ability of a cell to acquire the capability to grow in an environment missing an essential amino acid. In

these tests, a large number of cells are examined to demonstrate a significant increase in the frequencies of mutations that occur over the frequency of spontaneous mutations.

Chromosomal aberrations are large scale numerical or structural alterations in eukaryotic chromosomes including deletions (visualized as breaks), translocations (exchanges), non-disjunction (aneuploidy), and mitotic recombination. Chromosomal breakage is the classical end point in chromosomal aberration assays. Substances that induce structural changes in chromosomes, especially chromosome breaks, are referred to as "clastogens." To visualize chromosomes and chromosomal aberrations following *in vitro* or *in vivo* treatment with a substance, cells are arrested in metaphase, treated to swell the chromosomes, fixed, transferred to slides and stained. The first metaphase following treatment is the time at which the greatest number of cells with damaged chromosomes may be observed. The most frequently used test systems investigate changes in mammalian cells (such as Chinese hamster ovary or lung cells; human or rat lymphocytes; or human, rat or mouse bone marrow cells) following either *in vitro* or *in vivo* exposure to the test substance. The micronucleus test is a common *in vivo* assay that measures the frequency of micronuclei formation (i.e., chromosomal fragments) in polychromatic erythrocytes.

3.3.3.2 Summary of Mutagenicity Data

A summary of the mutagenicity information for the alkyl sulfides is presented in Table 6.

3.3.3.2.1 Bacterial Gene Mutation Assay

The bacterial reverse mutation assay considered adequate under the HPV Program has been conducted on four of the substances in this category: propanol/dodecylthio derivative (CAS # 67124-09-8), methyl propene derivative (CAS # 68511-50-2), trimethyl pentane derivative (CAS # 68515-88-8), and C15-C18 alkene derivative (CAS # 67762-55-4). These tests were conducted according to OECD Guidelines 471 and/or 472.

For two of the test substances, methyl propene derivative (CAS # 68511-50-2) and trimethyl pentane derivative (CAS # 68515-88-8), various strains of *S. typhimurium* and *E. coli* were tested with and without metabolic activation at doses that included and/or exceeded the limit dose of 5000 µg/plate. For the other two test substances, propanol/dodecylthio derivative (CAS # 67124-09-8) and the C15-C18 alkene derivative (CAS # 67762-55-4), the highest dose tested using various strains of *S. typhimurium* was 1 µl/plate (highest dose at which no precipitation was formed) with and without metabolic activation. All tested chemicals were negative for mutagenic activity, with and without metabolic activation.

3.3.3.2.2 *In vitro* Chromosomal Aberration Assay

An *in vitro* chromosomal aberration assay (using Chinese hamster ovary cells) was conducted for the propanol/dodecylthio derivative (CAS # 67124-09-8). This study is considered adequate under the HPV Program and was conducted in accordance with OECD Guideline 473. The results of this study, performed with and without metabolic activation of the test material, were negative for clastogenicity.

3.3.3.2.3 *In vivo* Chromosomal Aberration Assays

In vivo chromosomal aberration assays (using bone marrow cells from mice that were dosed by oral gavage or intraperitoneal injection) were conducted with methyl propene derivative (CAS # 68511-50-2), trimethyl pentane derivative (CAS # 68515-88-8), and an analog of the products in this class (CAS # 91770-97-4). The analog is a C12-C16 alkyl sulfide (CAS # 91770-97-4) bearing a methyl substituted side chain, with a structure very similar to the other alkyl sulfides in this group. These studies are considered adequate under the HPV Program and were conducted in accordance with OECD Guideline 474. All test substances were negative for clastogenicity. One of the test substances, the methyl propene derivative (CAS # 68511-50-2) was also tested in an *in vivo* micronucleus assay in rats via the dermal route of exposure. The results of this study were also negative.

Summary: Either bacterial gene mutation assays, *in vitro* chromosomal aberration assays, or *in vivo* chromosomal aberration assays have been conducted for four members of the category. Neither mutagenicity nor clastogenicity was exhibited by any of the substances in the referenced tests.. These tests demonstrate a lack of DNA reactivity either in the absence or presence of metabolic activation. The lack of predicted reactivity of the propanol/dodecylthio derivative (CAS # 67124-09-8) due to the stability of the aliphatic carbon-thio linkage overrides any apparent potential detrimental effects conferred by the relatively low molecular weight and electron localization effect of the hydroxyl moiety. These findings support the expectation, based on the similar structural features and physicochemical properties, that all the compounds in the alkyl sulfide category do not react with DNA and chromosomes at the cellular level. In addition, the consistency of available data regarding genotoxicity further supports treatment of all five alkyl sulfides as a chemical category within the HPV Program.

3.3.3.3 Data Assessment and Test Plan for Mutagenicity

Members of the alkyl sulfide group have undergone one or more test for mutagenicity and/or clastogenicity considered adequate under the HPV Program. In total, eight adequate mutagenicity studies have been conducted upon various members of the alkyl sulfide chemical category. The assays have included bacterial mutation, *in vitro* chromosomal aberration, and *in vivo* chromosomal aberration studies in rats and mice, which have involved four of the five members of the alkyl sulfide category. Additionally, an analog of one of the members of this category has also been evaluated in an *in vivo* chromosomal aberration study

with mice. The data consistently demonstrate a lack of mutagenicity. These findings support the expectation, based on the similar structural features and physicochemical properties, that the compounds in the alkyl sulfide category are also similar in their lack of mutagenic potential.

The HPV Challenge Program requires that a gene mutation and a chromosomal aberration test be performed, or bridged, to each member chemical of a category. Although mutagenicity testing considered to be adequate for the HPV Program has not been conducted on the decene derivative (CAS # 72162-15-3), the similarity of the structural features and physicochemical properties of the derivative is similar to those of the other members of this category. Thus, the decene derivative (CAS # 72162-15-3) is expected to exhibit genotoxicity results similar to those of the other alkyl sulfides in this category and additional testing of the decene derivative (CAS # 72162-15-3) is not needed. No additional mutagenicity testing is proposed for purposes of the HPV Challenge Program.

3.3.4 Repeated Dose Toxicity of the Alkyl Sulfide Category

3.3.4.1 Repeated Dose Toxicity Test Methodology

Repeated dose toxicity studies evaluate the effect(s) of repeated exposure to a chemical over a significant period of the life span of an animal. Chronic repeated dose toxicity studies are concerned with potential adverse effects upon exposure over the greater part of an organism's lifespan (e.g., one to two years in rodents). Subchronic repeated dose studies are also concerned with effects caused by exposure for an extended period, but not one that constitutes a significant portion of the expected lifespan. Typically, the exposure regimen in a subchronic study involves daily exposure (at least 5 consecutive days per week) for a period of at least 28 days or up to 90 days (i.e., 4 to 13 weeks). The dose levels evaluated are notably lower than the relatively high limit doses used in acute toxicity studies. In general, these studies are designed to assess systemic toxicity but the study protocol can be modified to incorporate evaluation of potential adverse reproductive and/or developmental effects.

3.3.4.1.1 Systemic Organ Studies

These studies provide information on systemic effects of a test substance in laboratory animals (rats, rabbits, or mice) when exposed via oral, dermal, or inhalation routes of administration over a limited duration (usually 28-90 days). A two-week recovery period (generally included in most study designs) following completion of the dosing or exposure period provides information on whether or not the effects seen during the exposure period are reversible. These studies are useful in identifying target organ(s), and they can be used in selecting dose levels for longer-term studies and for further refining safety criteria for human exposure. An indicator of the toxicity of a substance when tested in subchronic studies is the NOAEL (no observed adverse effect level). This measurement, usually expressed in mg/kg/day, defines the dose of test material that produced no significant toxicological

effects. Should the test material produce toxicity at the lowest dose tested (i.e., no defined NOAEL), the lowest dose that produced an adverse effect is presented as the LOAEL.

3.3.4.1.2 Reproductive/Developmental Studies

Reproductive and developmental toxicity studies generate information on the effects of a test substance on male and female reproductive performance such as gonadal function, mating behavior, conception, and development of the conceptus, parturition, and post-partum development of the offspring. Various study designs exist, but they all involve exposure to both male and female test animals beginning before mating. The rat is most often selected as the test species. The test substance is administered to males and females continuously at several graduated doses for at least two weeks prior to mating and until the animals are sacrificed. The males are treated for at least two more weeks. Male gonadal histopathology is carefully assessed at the end of the study. The females are treated through parturition and early lactation. The adult females and offspring are typically studied until termination on post-natal day 21, or sometimes earlier. In addition to providing data on fertility and reproduction, this study design provides information on the potential for prenatal exposure, and limited post-natal exposure to affect development. NOAEL or LOAEL are also applicable to these tests, with the exception that these values are derived from effects specific to reproduction or development.

The "toxicity to reproduction" requirement in the HPV Challenge Program can be met by conducting a reproductive/developmental toxicity screening test or adding a reproductive/developmental toxicity screening test to the repeated dose study (OECD 421 or OECD 422, respectively). The one-generation reproduction toxicity study (OECD 415) is a more comprehensive protocol for the study of the effect of a test material on reproduction and development that also meets the SIDS and the HPV Program requirements.

3.3.4.2 Summary of Repeated Dose Toxicity Data

A summary of the results from the repeated dose studies considered adequate for the HPV Program for alkyl sulfides is presented in Table 7.

3.3.4.2.1 Systemic Toxicity Tests

The following subchronic systemic toxicity studies are applicable to the chemicals in the alkyl sulfide group under the HPV Program:

- A 28-day (oral) repeated dose study in rats (with 2-week reversibility period). Dose levels of 100, 300, or 1000 mg/kg/day.
  (Test material: propanol/dodecylthio derivative (CAS # 67124-09-8)).

- A 13-week dermal administration to rats. Dose levels of 10, 50, 10, 250, 500, or 2000 mg/kg/day.

(Test material: methyl propene derivative (CAS # 68511-50-2))

- A 28-day subchronic dermal toxicity study in rabbits. Dose levels of 200 or 2000 mg/kg/day.
  (Test material: methyl propene derivative (CAS # 68511-50-2))

- A 21-day repeated dermal application study in rabbits. Dose levels of 140, 560, or 2240 mg/kg/day.
  (Test material: methyl propene derivative (CAS # 68511-50-2))

- A 28-day inhalation study in rats with a 3-week recovery period. Dose levels of 15, 50, or 150 mg/m^3.
  (Test material: trimethyl pentane derivative (CAS # 68515-88-8))

- A 28-day dermal toxicity study in rats. Dose level of 1,000 mg/kg/day.
  (Test material: trimethyl pentane derivative (CAS # 68515-88-8))

As noted above and in Table 7, four subchronic dermal toxicity studies (in rats or rabbits) have been conducted with either the methyl propene derivative (CAS # 68511-50-2) or trimethyl pentane derivative (CAS # 68515-88-8). The predominant effect noted was dermal irritation at the site of test material administration. Among the four subchronic dermal toxicity studies conducted for this category, the lowest reported NOEL was 50 mg/kg/day for a 13-week rat dermal study with methyl propene derivative (CAS # 68511-50-2).

A 28-day oral toxicity study in rats has been conducted with the propanol/dodecylthio derivative (CAS # 67124-09-8) and a 28-day inhalation study in rats has been conducted with the trimethyl pentane derivative (CAS # 68515-88-8). In both studies, gross and microscopic observations at study termination showed alterations in kidneys and liver which were similar in nature for both test materials. Gross observations included increased kidney and liver weights in both studies. Microscopic alterations in the kidneys were seen primarily in male rats and consisted of increased incidences of globular casts and the presence of hyaline droplets in the proximal tubule cells. Although the globular casts were also seen after the recovery period, no hyaline droplets were seen in the recovery animals, indicating that this change was reversible after cessation of test substance administration. This effect is considered to be an organ-, gender- and species-specific (i.e., kidney, male, rat) response that is commonly observed following repeated administration of long-chain aliphatic hydrocarbon-based materials.[13,14] The male rat is

[13] Halder, C.A., et al., Renal Toxicity of Gasoline and Related Petroleum Napthas in Male Rats, *Renal Effects of Petroleum Hydrocarbons*, M.A. Mehlman, C.P. Hemstreet, J.J. Thorpe and N.K. Weaver, eds., Princeton Scientific Publishers, Inc., Princeton, N.J., pp 73-88, 1984.

considered uniquely sensitive to such effects, and these effects are considered irrelevant to humans (cite!). Microscopic examination of the liver showed hypertrophy of hepatocytes in all animals at termination of the dosing period and in the high-dose animals at the end of the recovery period. These alterations represent a compensatory increase in the activity of hepatic metabolic processes in response to a xenobiotic challenge, and normally are not considered to be pathological.

Summary: Repeated-dose toxicity tests have been conducted on three of the alkyl sulfide compounds. The results of these studies consistently demonstrate relatively low systemic toxicity among the three tested substances including the marker compound, propanol/dodecylthio derivative (CAS # 67124-09-8). The effects on the liver that have been noted in these studies are considered to be adaptive responses. The effects on the kidney in male rats observed in these studies are not relevant to humans.

3.3.4.2.2 Reproductive/Developmental Toxicity

No reproductive or developmental toxicity data considered adequate under the HPV Program for the alkyl sulfide category are available.

3.3.4.3 Data Assessment and Test Plan for Repeated Dose Toxicity

3.3.4.3.1 Systemic Toxicity

Six repeated dose systemic toxicity studies involving two different animal species and three of the five members of the alkyl sulfide category have been conducted. The results of these repeated-dose toxicity studies in various laboratory animal models collectively show no significant adverse biological responses or specific toxicity relevant to humans resulting from exposure to chemicals in the alkyl sulfide group. The observed effects to the kidney were considered to be specific to the male rat and without relevance to humans, and the adaptive changes to the liver are not considered to be adverse effects. The structural features and physicochemical properties of the decene derivative (CAS # 72162-15-3) and the C15-C18 alkene derivative (CAS # 67762-55-4) are sufficiently similar to those of the other members of this category. Thus, these derivatives are expected to exhibit repeated dose toxicity results similar to those of the other alkyl sulfides in this category and additional testing is not needed. The repeated-dose toxicity of the category has been evaluated adequately with respect to all repeated dose toxicity HPV endpoints. No additional repeated dose toxicity testing is proposed for purposes of the HPV Challenge Program.

[14] Goldsworthy, Thomas L., et al., Potential Role of a-2 l-Globulin, Protein Droplet Accumulation and Cell Replication in the Renal Carcinogenicity of Rats Exposed to Trichloroethylene, Perchloroethylene and Pentachloroethane, *Toxicology and Applied Pharmacology*, Volume 96, pp. 367-379, 1988.

3.3.4.3.2 Reproductive/Developmental Toxicity

Although the alkyl sulfide category is well tested for other health effects and the data demonstrate similarity of toxicity across all members of the category, no reproductive/developmental studies considered to be adequate under the HPV Program are available to the HERTG. Based upon the previously discussed similarities in physicochemical properties of the alkyl sulfide category, in combination with the existing testing data confirming that the category exhibits low biological activity, it is reasonable to expect that exposure to alkyl sulfides will not result in reproductive/developmental effects.

However, given the absence of any reproductive/developmental testing data considered adequate under the HPV Program for this group, HERTG will conduct a 1-generation reproductive toxicity test upon the propanol/dodecylthio derivative (CAS # 67124-09-8). As discussed at the beginning of this section, it serves as a "marker chemical" that establishes the potential upper bound potential of mammalian toxicity for the entire group. Reproductive/developmental testing of only this marker chemical is consistent with EPA guidance in the document titled "Development of Chemical Categories in the HPV Challenge Program." This guidance, which states that "[I]n certain cases, such as where toxicity is low and no upward trend is expected, (emphasis added) extrapolation to the higher category members may be acceptable." The studies for all endpoints other than reproductive/developmental toxicity demonstrate that toxicity is relatively low among the members of this category. Thus, the decision to evaluate reproductive toxicity by testing the propanol/dodecylthio derivative (CAS # 67124-09-8) is both scientifically justified and consistent with EPA guidance.

If testing of the propanol/dodecylthio derivative (CAS # 67124-09-8) shows low reproductive/developmental toxicity, no further testing of the category will be considered necessary.

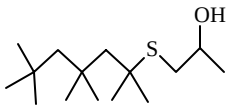
If the testing of the propanol/dodecylthio derivative (CAS # 67124-09-8) does yield test results that are positive or equivocal, the HERTG will evaluate the need for additional reproductive testing in this category. This stepwise approach to testing is scientifically justified, it is consistent with EPA guidance, and is preferred because it improves the quality of the subsequent research and reduces the unnecessary use of test animals.

Table 1. Members of the Alkyl Sulfide Category

Chemical Name	Simplified Chemical Name	CAS Number
2-propanol, 1-(tert-dodecylthio)-	propanol/dodecylthio derivative	67124-09-8
1-decene, sulfurized	decene derivative	72162-15-3
1-propene, 2-methyl-, sulfurized	methyl propene derivative	68511-50-2
Pentene, 2,4,4-trimethyl-, sulfurized	trimethyl pentane derivative	68515-88-8
Alkenes, C15-18 alpha-, sulfurized	C15-C18 alkene derivative	67762-55-4

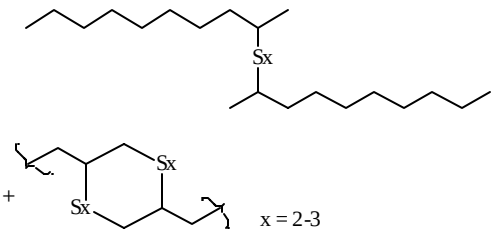
Figure 1. Chemical Structures

1. Propanol/dodecylthio derivative
CAS# 67124-09-8



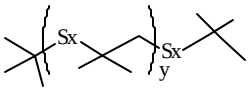
MW = 260

2. Decene derivative
CAS# 72162-15-3



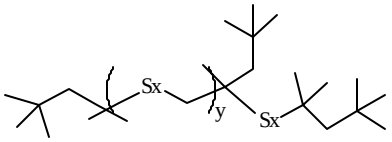
x = 2-3
MW = 344-472

3. Methyl propene derivative
CAS# 68511-50-2



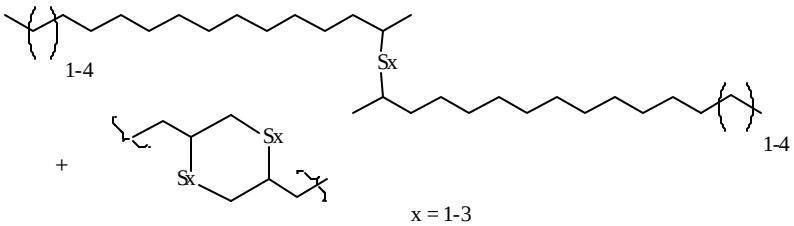
x = 1-5 y = 1-20
Mn = 800
MW = 320-2,300

4. Trimethyl pentane derivative
CAS# 68515-88-8



x = 4-5
y = 1
MW = 594-658

5. C15-C18 alkene derivative
CAS# 67762-55-4



x = 1-3
Mn = 520

MW = molecular weight
Mn = number average molecular weight

Table 2. Functional Group, Chemical Classes, and Hydrolytic Potential of Alkyl Sulfide Category Compounds

Members of the Alkyl Sulfide Category	Functional Group and Chemical Class	Potential for Hydrolysis
2-propanol, 1-(tert-dodecylthio)-	Alcohol	Low
	Alkane	Low
	Sulfide	Low
1-decene, sulfurized	Alkene	Low
	Alkane	Low
	Sulfide	Low
	Polysulfide	Low
1-propene, 2-methyl-, sulfurized	Alkene	Low
	Alkane	Low
	Sulfide	Low
	Polysulfide	Low
Pentene, 2,4,4-trimethyl-sulfurized	Alkene	Low
	Alkane	Low
	Sulfide	Low
	Polysulfide	Low
Alkenes, C15-18, alpha, sulfurized	Alkene	Low
	Alkane	Low
	Sulfide	Low
	Polysulfide	Low

Table 3. Evaluation of Environmental Fate Information

CHEMICAL	BIODEGRADABILITY	HYDROLYSIS	PHOTODEGRADATION
	PROPOSED TESTING OR INFORMATION FOR ENDPOINT	PROPOSED TESTING OR INFORMATION FOR ENDPOINT	PROPOSED TESTING OR INFORMATION FOR ENDPOINT
propanol/dodecylthio derivative (CAS # 67124-09-8)	Adequate data (5.9% biodegraded after 28 days)	No testing needed technical limitations[1]	AOPWIN Model Estimation
decene derivative (CAS # 72162-15-3)	Test	No testing needed technical limitations[1]	No testing needed Bridging
methyl propene derivative (CAS # 68511-50-2)	Adequate data (0.3% biodegraded after 28 days)	No testing needed technical limitations[1]	AOPWIN Model Estimation
trimethyl pentane derivative (CAS # 68515-88-8)	No testing needed Bridging	No testing needed technical limitations[1]	No testing needed Bridging
C15-C18 alkene derivative (CAS # 67762-55-4)	No testing needed Bridging	No testing needed technical Limitations[1]	AOPWIN Model Estimation

[1]See technical discussion included in hydrolysis section.

Table 4. Evaluation of Ecotoxicology Information

CHEMICAL	ACUTE TOXICITY TO FISH PROPOSED TESTING OR INFORMATION FOR ENDPOINT [96-HOUR LC50[1] (mg/L)]	ACUTE TOXICITY TO INVERTEBRATES PROPOSED TESTING OR INFORMATION FOR ENDPOINT [48-HOUR EC50[1] (mg/L)]	TOXICITY TO ALGAE PROPOSED TESTING OR INFORMATION FOR ENDPOINT [96-HOUR EC50[1] (mg/L)]
Propanol/dodecylthio derivative (CAS # 67124-09-8)	Test	Test	Test
Decene derivative (CAS # 72162-15-3)	No testing – bridging	No testing – bridging	No testing – bridging
Methyl propane derivative (CAS # 6811-50-2)	>1,000 (WAF[2], F) >10,000 (WSF[4], S)	>1,000 (WAF[3], D)	R >100 (WAF[3], P) B = 34 (WAF[3], P)
Trimethyl pentane derivative (CAS # 68515-88-8)	No testing – bridging	No testing – bridging	No testing – bridging
C15-C18 alkene derivative (CAS # 67762-55-4)	No testing – bridging	No testing – bridging	No testing – bridging

[1]Toxicity endpoints for the chemicals are expressed as median lethal concentration (LC_{50}) for fish and median effective concentration (EC_{50}) for *Daphnia* and algae. The EC/LC_{50} is defined as the concentration that adversely effects 50% of the test organisms exposed to the chemical during a specific time. The greater the EC/LC_{50} the lower the toxicity. See report text for information regarding differences between reported LC_{50} and EC_{50} values and lethal loading rate (LL_0) and effective loading (EC_0) values.

[2]WAF = Water accommodated fraction static renewal test.

[3]WAF = Water soluble fraction static non-renewal test.

[4]WSF= Water soluble fraction static renewal test.

F = fathead minnow, *Pimephales promelas.*

D = freshwater cladoceran, *Daphnia magna*.

P = freshwater algae *Pseudokirchneriella subcapitata* formerly called *Selenastrum capricornutum.*

S = sheepshead minnow, *Cyprinodon variegatus.*

R = growth rate

B = biomass

Table 5. Evaluation of Acute Toxicity Information

CHEMICAL	ACUTE TOXICITY PROPOSED TESTING OR INFORMATION FOR ENDPOINT
propanol/dodecylthio derivative (CAS # 67124-09-8)	Data Available • Oral – LD50 >5000 mg/kg (rat) • Dermal – LD50>2000 mg/kg (rabbit)
decene derivative (CAS # 72162-15-3)	No testing needed • bridging
methyl propene derivative (CAS # 68511-50-2)	Data Available • Oral – LD50 >5000 mg/kg (rat) • Oral – LD50 = 5.7 ml/kg (rat) Inhalation – LC50 >0.39 mg/L (rat) (highest dose tested)
trimethyl pentane derivative (CAS # 68515-88-8)	Data Available • Oral – LD50>5000 mg/kg (rat) • Dermal – LD50 >2000 mg/kg (rabbit) • Inhalation – LC50 = >5.6 mg/L (male rat); 2.17 mg/L (female rat) • Inhalation – LC50 >4.3 mg/L (male rat, mouse, guinea pig); <4.3 mg/L (female rat) (maximum attainable concentration) Inhalation – LC50 >4.3 (mouse, guinea pig)
C15-C18 alkene derivative (CAS # 67762-55-4)	Data Available Dermal – LD50>2000 mg/kg (rabbit)

Table 6. Evaluation of Mutagenicity Information

CHEMICAL	BACTERIAL GENE MUTATION ASSAY	*IN VITRO* CHROMOSOMAL ABERRATION ASSAY	*IN VIVO* CHROMOSOMAL ABERRATION ASSAY
	PROPOSED TESTING OR INFORMATION FOR ENDPOINT	**PROPOSED TESTING OR INFORMATION FOR ENDPOINT**	**PROPOSED TESTING OR INFORMATION FOR ENDPOINT**
propanol/dodecylthio derivative (CAS # 67124-09-8)	Adequate data Negative +/- S9	Adequate data Negative +/- S9	No testing needed • adequate data with *in vitro* test
decene derivative (CAS # 72162-15-3)	No testing needed • bridging	No testing needed • bridging	No testing needed • bridging
methyl propene derivative (CAS # 68511-50-2)	Adequate data Negative +/- S9	No testing needed • adequate data with *in vivo* test	Adequate data • Negative (mouse) Negative (rat)
trimethyl pentane derivative (CAS # 68515-88-8)	Adequate data Negative +/- S9	No testing needed • adequate data with *in vivo* test	Adequate data Negative (mouse)
C15-C18 alkene derivative (CAS # 67762-55-4)	Adequate data Negative +/- S9	No testing needed • adequate data with *in vivo* test	No testing needed • bridging
analog for C15-C18 alkene derivative (CAS # 91770-97-4)			Adequate data Negative (mouse)

Table 7. Evaluation of Repeated Dose Toxicity Information

CHEMICAL	REPEATED DOSE TOXICITY	REPRODUCTIVE/DEVELOPMENTAL TOXICITY
	PROPOSED TESTING OR INFORMATION FOR ENDPOINT	**PROPOSED TESTING OR INFORMATION FOR ENDPOINT**
propanol/dodecylthio derivative (CAS # 67124-09-8)	Adequate Data • STUDY: 28-Day Oral (rat) • Dose Levels: 100, 300, or 1000 mg/kg/day • Effects: ≥ 100 mg/kg/day: increased liver and kidney weights; globular casts; hyaline droplets in proximal tubules; hypertrophy of hepatocytes • NOEL: < 100 mg/kg/day (not defined in the report) Test 1-generation reproduction	Test 1-generation reproduction
decene derivative (CAS # 72162-15-3)	No testing needed • bridging	No testing needed • bridging
methyl propene derivative (CAS # 68511-50-2)	Adequate data • STUDY ONE: 13-Week Dermal (rat) • Dose Levels: 10, 50, 100, 250, 500, or 2000 mg/kg/day • Effects: ≥ 250 mg/kg/day: decreased body weight gain (males); decrease in RBC; increase in neutrophils; increase in spleen size and pigments in spleen; ≥ 100 mg/kg/day: increased production of WBC in spleen and bone marrow; ≥ 10% (100 mg/kg/day) concentration: dermal irritation • NOEL: 50 mg/kg/day (systemic)10% (dermal irritation) No testing needed • Adequate repeated dose data bridging repro data	No testing needed • Bridging

Table 7. Evaluation of Repeated Dose Toxicity Information (continued)

CHEMICAL	REPEATED DOSE TOXICITY	REPRODUCTIVE/DEVELOPMENTAL TOXICITY
	PROPOSED TESTING OR INFORMATION FOR ENDPOINT	**PROPOSED TESTING OR INFORMATION FOR ENDPOINT**
	• STUDY TWO: 28-Day Dermal (rabbit) • Dose Levels: 200 or 2000 mg/kg/day (intact and abraded skin) • Effects: ≥ 200 mg/kg/day: severe skin irritation; 2000 mg/kg/day: decrease in body weight and food consumption (males) NOEL: < 200 mg/kg/day (not defined in the report)	
methyl propene derivative (CAS # 68511-50-2)	• STUDY THREE: 21-Day Dermal (rabbit) • Dose Levels: 140, 560, or 2240 mg/kg/day • Effects: ≥ 140 mg/kg/day: severe erythema and slight to moderate edema. Epithelial hyperplasia of the skin in all treated animals. • NOEL: < 140 mg/kg/day (not defined in the report) No testing needed • Adequate repeated dose data bridging repro data	No testing needed Bridging

Table 7. Evaluation of Repeated Dose Toxicity Information (continued)

CHEMICAL	REPEATED DOSE TOXICITY	REPRODUCTIVE/DEVELOPMENTAL TOXICITY
	PROPOSED TESTING OR INFORMATION FOR ENDPOINT	**PROPOSED TESTING OR INFORMATION FOR ENDPOINT**
trimethyl pentane derivative (CAS # 68515-88-8)	Adequate data • STUDY ONE: 28-Day Inhalation (rat) • Dose Levels: 15, 50, or 150 mg/m^3 • Effects: ≥ 15 mg/ m^3: Trend toward lower body weight gain (all males and two highest doses in females); increased kidney weights (all males only); globular casts in cortico-medullary junction; hyaline droplets in proximal tubules (all males and recovery high dose males); increased liver weight (high-dose males and females and mid-dose males); 150 mg/L: decrease in hemoglobin concentration • NOEL: < 15 mg/ m^3 (not defined in the report) No testing needed • adequate repeated dose data bridging reproduction data	No testing needed • bridging

Table 7. Evaluation of Repeated Dose Toxicity Information (continued)

CHEMICAL	REPEATED DOSE TOXICITY	REPRODUCTIVE/DEVELOPMENTAL TOXICITY
	PROPOSED TESTING OR INFORMATION FOR ENDPOINT	PROPOSED TESTING OR INFORMATION FOR ENDPOINT
trimethyl pentane derivative (CAS # 68515-88-8)	• STUDY TWO: 28-Day Dermal (rat) • Dose Levels: 1000 mg/kg/day • Effects: Irritating to rat skin at 1000 mg/kg/day; Dermal irritation included well-defined erythema, scabbed areas, and dry flaking skin. Minimal to mild multifocal eschar and mild multifocal hemorrhage in the underlying dermis. • NOEL: < 1000 mg/kg/day No testing needed • adequate repeated dose data bridging reproduction data	No testing needed • bridging
C15-C18 alkene derivative (CAS # 67762-55-4)	No testing needed • bridging	No testing needed • bridging

ROBUST SUMMARY
ALKYL SULFIDE CATEGORY
CAS # 67762-55-4
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 67762-55-4
Chemical Name	Alkenes, C15-18 **alpha, sulfurized**
Remarks	This chemical is also referred to as C15-C18 alkene derivative in the HERTG's **Test Plan** for Alkyl **Sulfide** Category. For more information **on** the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	
Test **Type**	Acute **dermal** toxicity, single exposure
GLP (Y/N)	Y
Year (Study Performed)	1996;
Species/Strain	New **Zealand White** rabbits
Sex	Male and female
No. of animals/sex/dose	10/dose (5M, SF)
Vehicle	None, test article was doses as received.
Route of administration	Dermal, to clipped **intact, dorsal** skin
Remarks field for test conditions	**One dermal, semi-occluded patch** of **test article at 2,000 mg/kg was** applied to clipped dorsa **skin** of each animal. The patches were removed after 24 hours. All animals were observed daily for 14 **days** following test article administration.
Results	
Remarks	**No clinical signs** were observed during the study. Erythema and/or **edema** of the skin **at** application site were observed on **Day I in** some animals. There was an increase in mean body weight during the study. None of the animals died during the study. No visible lesions were observed in **any animal** at terminal necropsy.
Conclusions	**LD50 > 2000** mg/kg
Data Quality	**Reliable without restriction (Klimisch Code)**
References	This robust summary was prepared from an unpublished **study by an** individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-29-99

AR201-12549b2

ROBUST SUMMARY
ALKYL SULFIDE CATEGORY
CAS # 67762-55-4
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY N VITRO

Test Substance	
CAS #	CAS# 67762-55-4
Chemical Name	Alkenes, C15-18 alpha, sulfurized
Remarks	This chemical is also referred to as C15-C18 alkene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Designed to be in compliance with microbial mutagenicity testing as set forth by OECD 1981, EPA 1982, FDA 1993
Test Type	Reverse mutation assay
System of testing	Bacterial
GLP (Y/N)	Y, except analyses **were** not performed to verify the homogeneity, stability or accuracy of the test/control article preparation
Year (Study Performed)	1996
Species/Strain	Salmonella typhimurium - TA1535, TA1537, TA98, TA100 and TA102 Escherichia coli - WP2 uvrA
Metabolic activation	With and without
Concentrations	Prescreen, duplicate cultures: 50.0, 167, 500, 1670, and 5000 microgram/plate, plus Control Triplicate cultures: 50.0, 167, 500, l670,5000 and 10,000 micrograms/plate
Statistical methods	Statistical analyses are performed using the program developed by Snee and Irr (1981), with significance established at the 95% confidence limit.
Remarks field for test conditions	Test article was first evaluated in a prescreen using both liquid pre-. incubation and plate incorporation treatment conditions. Duplicate cultures of strains TA1537, TA100, an dWP2 uvrA were treated with article at doses of 50.0, 167, 500, 1670, and 5000 micrograms/plate, as well as the solvent control, in the absence of S9. The test article was found to be incompletely soluable (droplets were observed) at all doses. The article was next evaluated using both treatment conditions. Based upon the results of the prescreen, the article was evaluated in triplicate cultures in strains TA1535, TA1537, TA98, TA100, TA102, and WP2 uvrA in the presence and absence of S9 at doses of 50.0, 167, 500, 1670, 5000 and 10,000 micrograms/plate. Six doses of the article were evaluated in the event of unacceptable toxicity and/or insolubility at the highest dose levels evaluated in the mutation assay. The S9 mixture included 6% (v/v) Aroclor 1254-induced male Sprague-Dawley rat liver homogenate with the appropriate bugger and cofactors. The test article was again found to be incompletely soluble at all doses, under both treatment conditions. All Positive and negative controls were within acceptable ranges.

ROBUST SUMMARY
ALKYL SULFIDE CATEGORY
CAS # 67762-55-4
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VITRO

Results	
Remarks	In the prescreen, results indicated that the article was not toxic. In the following study, normal growth was observed in all tester strains at all doses evaluated with and without S9. Revertant frequencies for all doses of article in all tester strains, with and without S9 under both treatment conditions approximated or were less than those observed in the concurrent negative control cultures.
Conclusions	The results were negative in this study, using liquid pre-incubation and plate incorporation treatments.
Data Quality	Reliable with restriction (Klimisch Code). No analyses to verify test article preparation.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-29-99

AR 201-12549 b3

ROBUST SUMMARY
ALKYL SULFIDE CATEGORY
CAS # 67762-55-4
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VIVO

Test Substance	
CAS #	CAS # 91770-97-4
Chemical Name	Alkyl (C12-C16) sulfide.
Remarks	This chemical is an analog to the C15-C18 alkene derivative (CAS # 67762-55-4; Alkenes, C15-18 alpha, sulfurized) in the HERTG's Test Plan for Alkyl **Sulfide Category.** For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" **in** HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Method consistent with OECD 474 **and** EPA OPPTS 870.5395
Test Type	Mouse micronucleus test
GLP (Y/N)	Y
Year (Study Performed)	1996
Species	Mice
Strain	B6C3F1
Sex	Male
Route of administration	Intraperitoneal
Doses/concentrations	0, 500, 1000, and 2000 **mg/kg/day, plus** negative control (vehicle = corn oil) and positive control (= cyclophosphamide)
Exposure Period	**Three consecutive** days
Statistical methods	Statistical analysis was not performed on the frequency of micronucleated PCEs since test article animals had lower average numbers of micronucleated PCEs compared to controls.
Remarks field for test conditions	There was a range-finding phase of the study, which consisted of four groups of two male mice/group. Dose levels were 0, 500, 1000, and 2000. Groups of five mice each were dosed intraperitoneally with 0, 500, 1000, and 2000 mg/kg/day for three consecutive **days and then** sacrificed one day after the last **dose. The** positive control was administered as a single oral dose approximately 24 hours prior to sacrifice. Bone marrow cells were analyzed for the number of polychromatic erythrocytes (PCEs) which contained at least one micronucleus. A minimum of 2000 PCEs were analyzed from each animal from the vehicle control and from mice dosed with the test article. A minimum of 1000 PCEs was analyzed from each animal dosed with the positive control.

ROBUST SUMMARY
ALKYL SULFIDE CATEGORY
CAS # 67762-55-4
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VIVO

Results	
Remarks	The test article, when dosed to mice at 500, 1000 and 2000 mg/kg/day for three consecutive days did not induce an increase in the number of micronuclei. There was an indication of slight bone marrow cytotoxicity at the highest dose in the micronucleus phase. The decrease was statistically different from the vehicle control. This decrease was due to the lower percentage of PCEs for two animals. The responses obtained from the negative and positive control articles confirmed the reliability that the test system was capable of detecting compounds that induce micronuclei.
Conclusions	The test article did not cause an increase in micronuclei in developing erythrocytes in bone marrow from male B6F3C1 mice at the doses tested. There was a slight cytotoxic effect on developing erythrocytes at 2000 mg/kg/day, the maximum dose typically used in the mouse micronucleus phase.
Data Quality	Reliable without restrictions.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-29-99

AR201-12549b5

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
ECOTOXICITY ELEMENTS: TOXICITY TO AQUATIC PLANTS

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide **Category.** For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Test protocol followed US EPA Toxic Substances Control **Act** Test Guideline #797.1050 (1985, 1987), OECD Guideline for Testing of Chemicals #201 Alga, Growth Inhibition Test (1984).
Test Type	WAF static non-renewal **test**
GLP (Y/N)	v
Year (Study Performed)	1994
Species/Strain	Freshwater algae, ***Pseudokirchneriella subcapitata*** formerly called ***Selenastrum capricornutum***
Element basis (# of cells/ml)	~ 10,000 cells/ml
Exposure period/duration	96 hours
Analytical monitoring	Total organic carbon (TOC) measurements of initial test solutions **and** control (O-hour) and at test termination (96-h). EPA Method 415.1 (1979). Water samples were passed through 0.45 micron filter prior to TOC analysis.
Statistical Methods	
Remarks field for test conditions (fill as applicable)	Test Organisms: source - T.R Wilbury in-house culture originally purchased from the University of Texas at Austin algae collection. Test System: Individual WAFs were prepared for each test level and renewed daily. A measured weight of test material was added to a measured volume of dilution water (I-L) in a glass vessel **and stirred** for 24 hours. Stirring accomplished using a Telfon coated magnetic stir bar. Mixing speed adjusted such that a vortex formed between 30 to 50% of the distance to the bottom. Following the mixing period, the test solutions were allowed to stand for 1 hour before the water phase was removed. To avoid removing test material from the surface or bottom, a siphon was placed in the mixing vessel prior to addition of water and test substance with the lower end l-2 inches off the bottom. The siphoned water phase (i.e., WAF) was used for the aquatic toxicity test. A static test was conducted; i.e., there was no daily renewal of test solution. Three 100-mL replicates per treatment, inoculum -10,000 cells/ml. The 250-mL Erlenmeyer flasks were stoppered with foam plugs to reduce entry of dust, etc. During the test all treatment and control flasks were randomly placed on an orbital shaker adjusted to approximately 100 cycles per minute under constant light (24 hours/day). Daily cell counts were made

visually by means of direct microscopic examination with a hemocytometer. Cool-white fluorescent lights provided a light intensity of 47 to 50 uEin/m^2ec.

At the conclusion of the 96-h test a 0.5 mL subsample of test media from each 100 mg/L test flask was combined with 100 mL of fresh untreated alga media and incubated for up to 9 days or as soon as growth occurs. This was done to determine if growth inhibition was algistatic or algicidal.

Dilution Water: Sterile enriched alga growth media (US EPA, 1978, T.R. Wilbur-y SOP #6) adjusted to pH 7.5. Measured TOC and total suspended solids in fresh untreated alga media were <l.0 and <10 mg/L, respectively. Test temperature - 23.4 to 23.6 C, pH - 7.0 to 7.1 at O-hour and 8.6 to 10.2 after 96 hours. TOC measurements were only made on the lowest and highest test levels and control at the beginning and end of the test. TOC levels were <1.0 mg/L in the control and 1.0 mg/L WAF test level and 3 mg/L at 100 mg/L.

Test Levels: Control, 1.0, 5.0, 10, 50, 100 mg/L WAF loading rates.

Calculation of EL_{50}s and NOELRs: Moving average and probit methods (Stephan, 1983) were used to calculate EC_{50}s (i.e., EL_{50}s). A parametric one-way analysis of variance (ANOVA) and Dunnett's test were used to calculate the no-observed effect concentration (i.e., EL_0s) when data were normally distributed and a non-parametric Kruskal and Wallis test was used if data were not normally distributed.

Test Substance: No undissolved test material was seen on the surface of the test vessels during the entire aquatic toxicity test.

Reference Substance: No

Results

Measurements expressed as mg/L WAF loading rate:

	72-h EL_{50}	72-h NOELR[c]	96-h EL_{50}	96-h NOELR[c]
Cell Density:	26[a] (21-32)	5.0	34[b] (29-39)	10
Growth Rate:	>100	5.0	>100	10

[a]Moving average method. [b]Probitmethod.
Confidence limits in parentheses.
[c]Hypothesis analysis tests were used to determine NOELRs.

Re-growth of inhibited cultures from the 100 mg/L test level revealed the effect was algistatic rather than algicidal.

Remarks

Measured concentration: N/A

	Unit: mg/L WAF loading Element value: EL_{50} and NOELR (i.e., no-observable effect loading rate). - EL_{50}s and NOELRs reported as EC_{50}, and NOEC, respectively, although test results were based on WAF loading rates. - Test concentrations for the definitive test were not specified in a protocol amendment and the pH of the sterile media at the start of the test was 7.0 rather than 7.5. These deviations did not compromise the study.
Conclusions	Regrowth of inhibited cultures from the 100 mg/L test level revealed the effect was algistatic rather than algicidal.
Data Quality	Reliable without restrictions
References	Chemical Manufacturers Association, HERTG
Other	Updated: 12-27-99

AR201-12549b6

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
ECOTOXICITY ELEMENTS:
TOXICITY TO AQUATIC INVERTEBRATES (E.G., DAPHNIA)

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	U.S. EPA 797.1300 (1985,1987), OECD 202 (1984)
Test Type	Static acute toxicity test
GLP (Y/N)	Yes
Year (Study Performed)	1993
Species/Strain	Daphnia magna
Test details (static, semi-static, dosing rate, flow-through rate, etc.)	A static non-renewal test was conducted using water accommodated fractions (WAF) of the test material at 100, 300 and 1,000 mg/L loading rates. WAFs were prepared by adding a measured weight of the test material to a measured volume of the dilution water and stirring for 24 hours with a magnetic stir bar. The test solutions were allowed to stand for 1 hour before the water phase (WAF) was siphoned off.
Statistical Methods	Not conducted because there was greater than 50% survival in all test vessels.
Remarks field for test conditions (fill as applicable)	Test species: Juvenile daphnids, less than 24-hours old were produced from laboratory in-house culture Test conditions: Two 250-mL glass beakers that contained 200 ml of test solution were used per treatment. The 250-mL test vessels were loosely covered to reduce entry of dust, etc. Test temperature range: 20 ± 1°C Exposure vessel type: Dilution water: Filtered well water collected at Hampton, New Hampshire and adjusted to the appropriate hardness 168 to 172 mg/L as $CaCO_3$. The water was passed through activated carbon, a particle filter, and then an ultraviolet sterilizer and stored in a polyethylene tank where it was aerated. TOC levels were 2 mg/L at the beginning and end of the test, and 10 mg/L TSS at the beginning and <10 mg/L at the end of the test. Lighting: A 16 hour light and 8 hour dark photoperiod was maintained

	with cool-whit fluorescent lights with an intensity of 20 µEin^{-1}m^{-2}. Water chemistry: Dissolved oxygen - 8.2 to 8.7 mg/L; pH - 7.8 to 8.2; conductivity - 570 to 640 umhos/cm; temperature - 20.5 to 20.9°C. Element: Immobilization Test design: Control, 100, 300 & 1,000 mg/L WAF loading rates. 10 daphnids per replicate (20 per treatment). Method of calculating mean measured concentrations: not applicable Exposure period: 48 hours Analytical monitoring: Total organic carbon (TOC) measurements of initial test solutions and control (O-hour) and at test termination (48-h). TOC levels were 2 mg/L in the control, 3 mg/L at the 100 mg/L and 300 mg/L test levels; and 4 to 5 mg/L at the 1000 mg/L test vessel. TOC levels were not considered to be indicative of actual test material concentrations and results are therefore based on nominal loading rates
Results	Nominal concentrations: 48-hour and 24-hour EC50 = >1,000 mg/L (based on nominal loading rates). 48-hour and 24-hour NOEC = 1,000 mg/L
Remarks	Measured concentration: N/A Unit: mg/L EC50, EL50, LC0, LL0 at 24, 48 hours: 48-hour and 24-hour EC50 = >1,000 mg/L (based on nominal loading rates). 48-hour and 24-hour NOEC = 1,000 mg/L Statistical results: not applicable - Effect concentrations based on nominal loading rates - No immobilization seen at the highest test concentration of 1,000 mg/L (WAF) - Control response was satisfactory.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
ECOTOXICITY ELEMENTS:
TOXICITY TO AQUATIC INVERTEBRATES (E.G., DAPHNIA)

Conclusions	The WAFs of the test material were not toxic to daphnids at the concentrations tested. Ninety-five to 100% survival occurred at all test concentrations. No sublethal effects were noted during the test.
Data Quality	Reliable without restrictions
References	Chemical Manufacturers Association, HERTG Ward, T.J. (1993) Acute Toxicity of the Water Accommodated Fractions (WAFs) of CMA #613 to the Daphnid, ***Daphnia magna.*** T.R. Wilbur-y Study #9178-CMA/ESI-613.
Other	Updated: 12-21-99 This study is being submitted by the HERTG Panel of the Chemical Manufacturers Association.

AR201-12549b7

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
ECOTOXICITY ELEMENTS: ACUTE TOXICITY TO FISH

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is referred to as methyl propene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Test protocol followed US EPA Toxic Substances Control Act Test Guideline #797.1400 (1985), OECD Guideline for Testing of Chemicals #203, Fish Acute Toxicity Test (1984).
Test Type	WAF static renewal test
GLP (Y/N)	Y
Year (Study Performed)	1993
Species/Strain	Fathead minnow ***(Pimephales promelas)***
Analytical Monitoring	Total organic carbon (TOC) measurements of initial test solutions and control (O-hour) and after one day on test (24-h) before daily renewal of fresh test solution.
Exposure Period (unit)	96 hours
Statistical methods	Statistical analysis of survival data not warranted.
Remarks field for test conditions, (fill as applicable)	Test Organisms: source - Aquatic Research Organisms, Hampton, New Hampshire, age -juvenile, total length - 25 mm average (longest fish not more than twice the shortest fish), wet weight - 0.1 g average (no range reported). Loading - <0.5 g biomass/l, Pretreatment - none, fish held for a minimum of 14 days before testing. No feeding during the test. Test System: Individual WAFs were prepared for each test level and renewed daily. A measured weight of test material was added to a measured volume of dilution water (30-L) in a glass vessel and stirred for 24 hours. Stirring accomplished using a Telfon coated magnetic stir bar. Mixing speed adjusted such that a vortex formed between 30 to 50% of the distance to the bottom. Following the mixing period, the test solutions were allowed to stand for 1 hour before the water phase was removed. To avoid removing test material from the surface or bottom, a siphon was placed in the mixing vessel prior to addition of water and test substance with the lower end 1-2 inches off the bottom. The siphoned water phase (i.e., WAF) was used in the aquatic toxicity test. About 80% of the solution in each test level was renewed daily after 24, 48, and 72 hours. Two 15-L replicates per treatment, 10 fish per replicate (20 per treatment). Test vessels loosely covered to reduce entry of dust, etc.

	Dilution Water: Filtered well water collected at Hampton, New Hampshire and adjusted to the appropriate hardness of 176 mg/L as $CaCO_3$. The water was passed through activated carbon, a particle filter, and then an ultraviolet sterilizer, and then it was stored in a polyethylene tank where it was aerated. The water was characterized as moderately hard water. Alkalinity not reported. Dissolved oxygen - 7.1 to 8.7 mg/L, pH - 7.8 to 8.5, conductivity - 570 to 650 umhos/cm, temperature - 21.4 to 22.8 C. TOC levels were between 2 to 3 mg/L in the control, 3 mg/L at 100 mg/L test level, between 3 to 4 mg/L at 300 mg/L test level and 5 mg/L at 1,000 mg/L test concentration level. Test Levels: Control, 100, 300 & 1,000 mg/L WAF loading rates. Test Findings: No mortality was observed in all treatments and the control throughout the entire test and no signs of toxicity were noted in all treatments throughout 72 hours. At 96 hours, all 20 fish in the 1,000 mg/L test level were lethargic and exhibited erratic swimming, but no signs of toxicity were observed in the lower test levels and control. Calculation of LL_{50}s: Statistical analysis of survival data not warranted. Test Substance: No undissolved test material was seen on the surface of the test vessels during the entire aquatic toxicity test. Reference Substance: No
Results	Nominal concentrations: 96-h LL_{50} >1,000 mg/L. 96-h LL_0 = 300 mg/L. No mortality at 1,000 mg/L but at 96 hours all fish were lethargic and exhibited erratic swimming. TOC measurements at 1,000 mg/L were 5 mg/L compared to 2 to 3 mg/L in the control.
Remarks	Measured concentration: n/a Unit: mg/L LC50, LC0, LL50 or LL0 at 48, 72, 96 hours: LL_{50} and LL_0 reported as LC_{50} and NOEC, respectively, although test results were based on WAF loading rates. Statistical results: Statistical analysis of survival data not warranted. Other: ✍ The TOC in dilution water at the beginning and end of the test was greater than 2 mg/L rather than <2 mg/L. It could not be verified that water samples were passed through a 0.45 micron filter prior

	to TOC analysis, that the test vessels were covered, or that the continuous temperature measurement was made in a control vessel during the study. But, these deviations did not compromise the study.
Conclusions	No mortality was observed in all treatments and the control throughout the entire test and no signs of toxicity were noted in all treatments throughout 72 hours. At 96 hours, all 20 fish in the 1,000 mg/L test level were lethargic and exhibited erratic swimming, but no signs of toxicity were observed in the lower test levels and control.
Data Quality	Reliable without restrictions
References	Chemical Manufacturers Association, HERTG Ward, T. J. (1993) Acute Toxicity of The Water Accommodated Fractions (WAFs) of CMA 613 to The Fathead Minnow, *Pimephales promelas.* T.R. Wilbury Study #9176-CMA/ESI-613.
Other	Updated: 12-21-99

AR201-12549b8

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
ECOTOXICITY ELEMENTS: ACUTE TOXICITY TO FISH

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is referred to as methyl propene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Test protocol followed OECD Guideline for Testing of Chemicals #203, Fish Acute Toxicity Test (1984).
Test Type	WSF static renewal test; a one level screening test
GLP (Y/N)	Y
Year ***(Study*** Performed)	1986
Species/Strain	Sheepshead minnow ***(Cyprinodon variegatus)***
Analytical Monitoring	Total organic carbon (TOC) measurements of each freshly prepared test solution and control and after 24-h on test just before daily renewal with fresh test solution.
Exposure Period (unit)	96 hours
Statistical methods	Statistical analysis of survival data not warranted.
Remarks field for test conditions (fill as applicable)	Test Organisms: source - a commercial supplier in New Hampshire, age - 24 to 29 days old, total length - 20.3 mm average (range 13 to 30 mm; n=18), wet weight- 0.16 g average (range 0.03 to 0.039 g; n = 18). Loading - 0.32 g biomass/L, Pretreatment - none, fish held for a minimum of 20 days before testing. No feeding during the test. Test System: Individual WSFs were prepared for each daily renewal of the 10,000 mg/L test level. A measured weight of test material was added to a measured volume of dilution water (1 S-L) in a glass vessel and stirred for 16 to 24 hours. Stirring accomplished using a Telfon coated magnetic stir bar. Mixing speed adjusted such that a vortex formed between 30 to 50% of the distance to the bottom. Following the mixing period, the test solutions were allowed to stand for 2 hours before the water phase was removed. To avoid removing test material from the surface or bottom, a siphon was placed in the mixing vessel prior to addition of water and test substance with the lower end approximately midway between bottom and surface. The siphoned water phase, designated water soluble fraction (WSF), was used for the aquatic toxicity test. About 90% of the test solution in each test vessel was renewed daily after 24, 48, and 72 hours. Two 5-L replicates per treatmeat, IO fish per replicate (20 per treatment). Test vessels were loosely covered to reduce entry of dust, etc. Dilution Water: Natural seawater collected from Cape Cod Canal,

	Bourne, Massachusetts which derives water from Buzzards Bay or Massachusetts Bay. The water was filtered through OS-micron polypropylene core filter and activated carbon, then stored for 1 to 4 days prior to use while being constantly aerated. During storage the water had a salinity of 32 to 33 ppt and pH of 7.7 to 7.8. During the test: dissolved oxygen - 5.6 mg/L to above 100% saturation (7.5 mg/L), pH - 6.9 to 8.0, salinity - 32, temperature - 20 to 22 C. Mean measured TOC levels in the control and 1,000 mg/L WSF test level were 2.7 mg/L, (range 1.2 to 6.0) and 4.5 mg/L, (range 3.0 to 5.6), respectively Test Levels: Control & 10,000 mg/L, WSF loading rate. Test Findings: No mortality or signs of toxicity were noted in the 10,000 WSF test level and the control throughout the entire test. Calculation of LL_{50}s: Statistical analysis of survival data not warranted. Test Substance: No undissolved test material was seen on the surface of the test vessels during the entire aquatic toxicity test. Reference Substance: Sodium lauryl sulfate (SLS). The 96-h LC_{50} was 1.2 mg/L. No information provided on method of calculation.
Results	Nominal concentrations: 96-h LL_{50} >10,000 mg/L. 96-h LL_0 = 10,000 mg/L (no mortality or toxic signs noted). Mean measured TOC in the 10,000 mg/L WSF test level was 4.5 mg/L compared to 2.7 mg/L in the control.
Remarks	Measured concentration: n/a Unit: mg/L LC50, LC0, LL50 or LL0 at 48, 72, 96 hours: LL_{50} and LL_0 reported as LC_{50} and NOEC, respectively, although test results were based on WSF loading rate. Statistical results: Statistical analysis of survival data not warranted. Other:

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
ECOTOXICITY ELEMENTS: ACUTE TOXICITY TO FISH

Conclusions	No mortality or signs of toxicity were noted in the 10,000 WSF test level and the control throughout the entire test.
Data Quality	Reliable without restrictions
References	Chemical Manufacturers Association, HERTG Nicholson, R.B. (1986) Acute toxicity of CMA Test Material Code 525 to Sheepshead Minnow, ***Cyprinodon variegatus.*** Springborn Bionomics Study #10823-0186-6100-500-525, Report #BW-86-04-***2004.***
Other	Updated: 12-21-99

AR201-12549b9

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information ***on*** the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Experimental; modified OECD guideline 403
Test Type	Acute inhalation toxicity (4 hours)
GLP (Y/N)	Y
Year **(Study** Performed)	1990
Species/Strain	Rat; Sprague-Dawley
Sex	Male & Female (9 weeks old at initiation of study)
No. of animals/sex/dose	20 (10M, 10F/group)
Vehicle	Not applicable
Route of administration	Whole body inhalation of vapor from sample heated to 200°C
Remarks field for test conditions	Rats were exposed to 0.07 or 0.39 mg/l vapor for a single 4 hour whole body exposure. Sham control rats were placed in inhalation chambers in room air. One half of rats (5M, 5F) from each group were sacrificed 24 hour *post* exposure; others maintained for 2 weeks recovery. Vapors from the methyl propene derivative were generated in a counter current generator and delivered into the exposure chamber. Air samples were pulled through glass fiber filters to verify that particles were not present and animals were being exposed to pure vapor. Vapors were analyzed by GC to quantitate chamber concentrations using octane as a standard; qualitative analyses were performed by GC/MS. Animals were observed in the chambers and *post* exposure for clinical signs of toxicity. Body weights were taken and selected organs weighed at necropsy. Histopathology on liver, kidney lungs, nasal turbinates, tracheobronchial lymph nodes.
Results	LC50 > 0.39 mg/l
Remarks	No mortality and minimal toxicity was observed. Abnormal clinical signs occurring during and immediately post exposure included oral and ocular discharge, shallow respiration (some high dose rats only) and decreased response to stimuli. No abnormal treatment-related clinical signs were observed from I hour post exposure to the end of the recovery period, Group body weights and weights of liver, kidney and lungs were unaffected by exposure. No treatment related microscopic lesions were observed in the 5 organs examined.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: ACUTE TOXICITY

Conclusions	LC50 > 0.39 mg/l of a vapor generated from the starting material at 200°C, not from the whole methyl propene derivative. Vapor approximates conditions of workplace exposure.
Data Quality	Reliable with restrictions. GLP compliant Study performed with two dose groups and is not a limit test. Physical and chemical properties provided with exception of structural formula, purity of starting material.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-29-99

AR 201-12549 b10

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propane, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Experimental
Test Type	Acute oral toxicity (LD50)
GLP (Y/N)	**N**
Year (Study Performed)	1970
Species/Strain	Rat; Sherman/Wistar
Sex	Male, young adult
No. of animals/sex/dose	5
Vehicle	None: administered undiluted
Route of administration	Oral gavage
Remarks field for test conditions	Rats fasted 24 hours prior to dosing; Test material administered by gavage in a single oral dose at concentrations of 2.0, 4.0, 8.0, 16.0 or 32.0 ml/kg. Animals observed for 14 days postdosing for signs of toxicity or mortality. Body weights were not taken; gross necropsies and histopathology were not performed
Results	5.7 ml/kg; 19/20 confidence limits 4-8 ml/kg
Remarks	**No** deaths were observed at 2.0 or 4.0 ml/kg; at 8 ml/kg 4/5 dead at day 1 post dosing, 1/5 dead at day 2; S/S rats dead at day 1 in groups given 16.0 or 32.0 ml/kg. No data presented on parameters other then mortality.
Conclusions	LD5O = 5.7 ml/kg (male rats)
Data Quality	Reliable with restrictions State of the art LD5O oral toxicity study for 1970, multi-dose, calculated LD50 with confidence limits. Non-GLP, details of study design and observations during study not presented
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	updated: 12-29-99

AR 201-12549611

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methy propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information **on the** chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Litchfield and Wilcoxen (J. Pharm. & Exp. Therap. 96:99, **1949)**
Test Type	Acute oral toxicity (LD50)
GLP (Y/N)	**N**
Year (Study Performed)	1979
Species/Strain	Rat/Wister
Sex	Male
No. of animals/sex/dose	40 (4 groups of 10)
Vehicle	None: administered undiluted
Route of administration	***Oral gavage***
Remarks field for test conditions	Rats fasted for 24 hours prior to dosing; Test material administered by gavage in a single oral dose at concentrations of 5.0, 7.12, 10.14, and 14.43 g/kg. Animals observed 3-4 hours after dosing and once daily for 14 days post-dosing. Mortality, toxicity, and pharmacological effects were recorded for each animal. Body weights were recorded pretest. At the end of 14 days, all survivors were sacrificed and all animals, including those which died during the course of the study, were examined for gross pathology.
Results	LC50 = (6.8 - 10.9)g/kg
Remarks	At 5.0 g/kg 2/10 died (1 at day I post dose and l at day 13 post dose); at 7.12 g/kg **4/10 died** (3 at day 2 post dose and 1 at day 6 post dose); at 10.14 g/kg 6/10 died at day 2 post dose; at 14.43 g/kg 10/10 died (I at day 1 post dose and 9 at day 2 post dose). Toxicity was observed for the following doses: at 5.0 g/kg lethargy, **ataxia,** ptosis, piloerection and flaccid muscle tone were noted in 5 or more animals. Isolated instances of diarrhea, chromorhinorrhea, chromodacryorrhea and **tachypnea were** also noted; at 7.12 g/kg lethargy, diarrhea, piloerection, ptosis, chromodacryorrhea, ataxia, and chromorhinorrhea were noted in 5 or more animals. Isolated instances of tachypnea, **respiratory** noise and prostration were also noted; At 10.14 g/kg **lethargy,** diarrhea, chromorhinorrhea, ptosis, piloerection, chromodacryorrhea and ataxia were noted in 5 or more animals. Isolated instances of emaciation, prostration and hyperactivity were also noted; at 14.43 g/kg lethargy, ataxia, ptosis, diarrhea, chromorhinorrhea and piloerection were noted in 5 or more animals. Overall body weights increased slightly in the 5.0, 7.12, and 10.14

	g/kg doses. No body weights were reported for the 14.43 g/kg dosed animals. Necropsy was also performed.
Conclusions	LD50 = 8.6 g/kg (male rats)
Data Quality	Reliable with restrictions State of the art LD50 oral toxicity study for 1979, multidose, calculated LD50, no reference to confidence limits. Non-GLP, details of the study design and other observations not presented. Used 16 CFR 1500.3(c) (2) (I) for toxicity definitions.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-28-99

AR 201-12549612

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS#68511-50-2
ENVIRONMENTAL PATE AND PATHWAY ELEMENTS: BIODEGRADATION

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 'Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 301B, Ready Biodegradability, Modified Sturm Test
Test Type (aerobic/anaerobic)	Aerobic
GLP (Y/N)	Y
Year (Study Performed)	1996
Contact time (units)	28 days
Inoculum	Domestic sewage sludge plus soil

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS: BIODEGRADATION

Remarks for test conditions	Inoculum: Sludge from domestic WWTP used at 30 mg dry solids/L; soil from forest area used at 0.1 g/L Conc of test chemical: Test chemical added directly to test vessels at 13.3 mg C/L (28.6 mg/L: CAS# 68511-50-2). No preacclimation was used. Temp of incubation: 23 - 24°C Dosing procedure: Neat test chemical added by micropipettor to culture medium in vessels immediately prior to addition of sewage and soil inocula Sampling: Days 1, 3, 6, 10, 14, 21, 29 (after acidification on d 28) Controls: Yes (blank and positive controls used per guideline); toxicity control not used. Positive Control was Benzoic acid (Na salt) at 20 mg C/L Analytical method: Titration of residual Ba(OH)2 in trapping solution, using HCl Method of calculating measured concentrations: N/A; CO2 evolution and % biodegradation were calculated using the average of duplicate blank-corrected titration volumes at each titration interval Other: - The % biodegradation value reported is slightly inflated by the use of zero titration volume rather than negative volume when corrected for blanks; however, comparison of titration volumes for the test chemical and blank show them to be very similar, so inhibition of inoculum is not suspected.
Results	Not Readily Biodegradable
Degradation % after time	0.3% in 28 days
Kinetic (for sample, positive and negative controls)	$t_{1/2}$ for Positive Control was <10 d
Breakdown Products (Y/N) If yes describe breakdown products	N

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS #68511-50-2
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS: BIODEGRADATION

Conclusions	Not Readily Biodegradable; biodegradation was essentially zero
Data Quality	Reliable without restrictions
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12/29/99

AR 201-12549613

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VIVO

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	modified OPPTS 870.5395
Test Type	Mammalian bone marrow erythrocyte micronucleus test; adjunct to 13 week dermal subchronic toxicity study
GLP (Y/N)	Y
Year (Study Performed)	1987
Species	Rat
Strain	Sprague Dawley (Tac:N[SD]fBR)
Sex	Male **and** female
Route of administration	dermal to shaved **skin** of backs
Doses/concentrations	500 and 2000 mg/kg/day undiluted test material; 500 mg/kg/day diluted 50%w/v in 100" mineral oil base stock 10 (5M,5F/group): 3 treatment groups, 1 untreated controls vehicle = mineral oil (100" solvent refined naphthenic base stock) density 0.88 g/ml
Exposure Period	5 days/week for 13 weeks
Statistical methods	SAS ANOVA and ANOVA F test; Tukey's Studentized Range test and Scheffe's test; SAS General Linear Model, a studentized linear regression analysis to determine dose responsiveness. Statistical analyses compared test **values** to negative control data; a significant increase in micronuclei is an indicator of clastogenic activity by the test material
Remarks field for test conditions	Age at initiation: 7 weeks old following 2 weeks acclimation Methyl propene derivative was applied to the clipped backs of groups of 20 Sprague Dawley rats (10M,10F) 5 days per week for 13 weeks at dose levels of 0, 500 or 2000 mg/kg/day undiluted or 500 mg/kg diluted (50% w/v) in 100" mineral oil base stock. Rats were fitted with Elizabeth collars to minimize ingestion of test material, which was left uncovered on the skin. At termination of the 13 week subchronic study, approximately 24 hours after the final dermal administration, bone marrow was harvested from femurs of the first 5 rats/sex/group necropsied. Three bone marrow slides were prepared for each animal. Slides were stained with acridine orange and scored under a

	fluorescence microscope. All slides were randomized by a computer-generated random numbers table so that the cytogeneticist was unaware of what dose group any individual slide was from. Immature red blood cells (polychromatic erythrocytes, PCE) and mature red blood cells (normochromatic erythrocytes, NCE) were evaluated for toxicity and the presence of micronuclei. The ratio of PCE to NCE per the first 1000 erythrocytes counted was calculated to determine cytotoxicity if any. At least 1000 PCE and 1000 NCE were scored for the presence of micronuclei.
Results	
Remarks	Methyl propene derivative undiluted (500 or 2000 mg/kg/day) and methyl propene derivative (500 mg/kg/day) 50% w/v in 100" mineral oil base stock were not cytototoxic to red blood cell formation. These test materials did not induce any statistically significant increase in the formation of micronucleated PCEs or NCEs in bone marrow red blood cells of male or female rats exposed dermally for 13 weeks.
Conclusions	Methyl propene derivative does not cause chromosome damage in rats following regular and prolonged dermal exposure in this test system. NOEL= 2000 mg/kg/day
Data Quality	Reliable with restrictions. Study does not include a positive control. Samples were collected only once approximately 24 hours after the final dose. Information on test material composition, purity and stability are not part of this report but are referred to the 13 week subchronic study report.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Undated: 12-29-99

AR 201-12549 b14

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VIVO

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 474.
Test Type	Mammalian bone marrow erythrocyte micronucleus test
GLP (Y/N)	Y
Year (Study Performed)	1989
Species	Mouse
Strain	B6C3F1
Sex	Male and female
Route of administration	Intraperitoneal
Doses/concentrations	Single injection of 3.5 g/kg test material, 50 mg/kg cyclophosphamide in saline, or methylcellulose vehicle alone (vehicle = hydroxypropyl methylce1lulose (Methocel K4M Premium - Dow Chemical)) 15 animals (5M, SF/dose/sample interval); Positive control (5M,5F)
Exposure Period	Single dose
Statistical methods	Normal test for equality of proportion (one-tailed). Because of multiplicity of comparisons, a Dunnett adjustment was made.
Remarks field for test conditions	Young male and female mice were treated with a single intraperitoneal injection of 3.5 g/kg test material, 50 mg/kg cyclophosphamide in saline, or methylcellulose vehicle alone. Dose had been determined in a preliminary toxicity test to identified MTD for this study. Animals were sacrificed and femurs removed at 24, 48 or 72 hours post dosing (5M, 5F per interval) for test material and negative control, and at 24 hours postdosing only for cyclophosphamide. Bone marrow smears were prepared and immature red blood cells (polychromatic erythrocytes, PCEs) and mature red blood cells (normochromatic erythrocytes, NCEs) were evaluated for toxicity and the presence of micronuclei. Slides were stained with acridine orange and scored under a fluorescence microscope. Slides from all dose groups were sorted by a computerized random number system and the cytogeneticist was unaware of what dose group any individual slide was from. The ratio of PCE or NCE per the first 1000 erythrocytes counted was calculated to determine cytotoxicity if any.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VIVO

Results	
Remarks	In the preliminary toxicity test (2M, 2F/group) all mice died at 5.0 g/kg and all survived at 3.5 g/kg with no cytotoxicity in bone marrow cells 24 hours after injection. Data from the full study demonstrate that the frequency of mironucleated PCEs in femoral bone marrow for males and females treated with the test material was not significantly elevated ($p<0.05$) when compared to negative controls for groups sampled at 24, 48 or 72 hours postinjection. Results from both sexes combined demonstrate the same results. Cyclophosphamide, the positive control material did induce statistically significant increases in micronucleated PCEs in all animals demonstrating that a valid study was performed.
Conclusions	Methyl propene derivative administered IP at 3.5 g/kg body weight did not induce the formation of micronuclei in PCEs in male or female mice at any time interval and is not considered clastogenic in this test system.
Data Quality	Reliable without restrictions. Guideline study.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-29-99

AR 201-12549 b15

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 'Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Reference for study design: Federal Register, Volume 43, Number 163
Test Type	28 Day Subchronic Dermal Toxicity Study
GLP (Y/N)	Y
Year (Study Performed)	1982
Species	Albino Rabbits
Strain	
Route of administration	Dermal to shaved dorsal trunk area of abraded or intact skin
Duration of test	
Doses/concentration levels	Group 1: 200 mg/kg/day undiluted test material Group 2: 2000 mg/kg/day undiluted test material 36 animals tested: (6M,6F/group): 2 treatment groups, I untreated control
Sex	Male and female
Exposure period	
Frequency of treatment	
Control group and treatment	1 untreated control group (6M/6F)
Post exposure observation period	
Statistical methods	All data was submitted to analysis of variance (method: Statisticat Analysis System, SAS Statistical Institute 1979) followed by evaluation using the Newman-Keuls method for all significant dose differences.
Remarks field for test conditions	Age at initiation of treatment: Not specified following 1 week acclimation. Study was designed to evaluate the subchronic toxicity of the test material when applied **dermally.** Methyl propene derivative was applied to the shaved dorsal trunk area (approximately 10% of the body surface) of 2 groups of 12 albino rabbits (6M,6F) 5 days per week for 4 weeks at dose levels of 200 or 2000 **mg/kg/day** of undiluted test material on the same test schedule. Half the animals in each group were abraded once per week throughout the **study.** The abrasion

	penetrated the stratum corneum but did not cause bleeding. The treated skin was occluded for at least 6 hours daily and the trunk of each animal covered with an impervious material. One untreated shaved control group of 6 animals (3 intact, 3 abraded) was included in the study. Assessments for local and systemic effects included clinical observations, skin irritation scoring 5 days per week, body weights (pretest, every 3 to 4 days during testing, termination), hematology, serum chemistry and urinalysis at pretest and termination, and gross necropsy evaluations at study termination.
Results	
Remarks	One male rabbit death at the higher dose level. Body weight gains in control (0.5 to 1.0 kg) and lower dose group (0.2 to 1.0 kg). A trend of weight loss and food consumption among the high dose males in the latter half of the study. Weight gain in 5 of 6 high dose females. Treatment with the test material caused severe skin irritation at both doses. Abrasion of skin increased the degree of irritation at the low dose level. No irritation was observed in the control group. Urinalysis values were normal for all groups. The low dose group showed an increase in chloride and a decrease in albumin. The high dose group showed decreased alkaline phosphatase and an increase in chloride and globulin. Hematology showed no trends in the control and low dose groups while monocyte determinations were significantly different (increased) in the high dose group. Gross and histopathological examination of tissues did not reveal any pattern of changes attributable to dermal contact with the test material. At autopsy one animal in the control group was found to be female instead of male and one animal in the low dose group was found to be male instead of female. Statistical evaluation including and excluding these two animals showed no significant differences. The hematological and clinical chemistry data do not suggest a consistent trend indicative of a response to the test compound
Conclusions	A NOAEL was not established in this study. A LOEL was not established in this study. No minimally irritating concentration was identified by this study.
Data Quality	Reliable with restrictions. Animal ages were not included in the report. Uneven sex distribution. Clinical behavior determinations beyond that of morbidity were not included in the report.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-28-99

AR 201-12549616

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	comparable to OPPTS 870.3250
Test Type	Thirteen **week** dermal subchronic toxicity study
GLP (Y/N)	Y
Year (Study Performed)	1989
Species	Rat
Strain	Sprague Dawley (Tac:N[SD]fBR)
Route of administration	Dermal to shaved skin of backs
Duration of test	5 days/week for 13 weeks
Doses/concentration levels	Part 1:500 and 2000 mg/kg/day undiluted test material; 500 mg/kg/day diluted 50%w/v in 100" mineral oil base stock Part 2: 500, 250, 100, 50, 10 mg/kg/day diluted in mineral oil base stock at concentrations of 25, 10 and 5% w/v respectively 20 (10M, 10F/group): 8 treatment groups, 1 vehicle control, 2 untreated controls
sex	Male and Female
Exposure period	
Frequency of treatment	
Control group and treatment	1 vehicle control,2 untreated controls Vehicle: mineral oil (100" solvent **refined** naphthenic base stock) density 0.88 g/ml
Post exposure observation period	
Statistical methods	Analysis of Variance followed by multiple range tests, Serum chemistry and hematology data were evaluated using the F test for ANOVA and Student-Newman-Keuls multiple comparison test.
Remarks field for test conditions	Age at initiation: 7 weeks old following 2 weeks acclimation Study was designed to identify inherent toxicity of test material and to determine whether dilution in a mineral oil carrier would alter toxicity. **Methyl** propene derivative was applied to the clipped backs of groups of 20 Sprague Dawley rats (10M,10F) 5 days per week for 13 weeks at dose levels of 500 or 2000 mg/kg/day undiluted or diluted in 100" mineral oil at dose levels of 500 250 100 50 or 10 mg/kg/day on the

	same schedule. Rats were fitted with Elizabeth collars to minimize ingestion of test material, which was left uncovered on the skin. One vehicle and 2 untreated shaved control groups were included in the study. Assessments for toxic response included daily clinical observations, weekly skin irritation scoring, weekly body weights and terminal organ weights, hematology, serum chemistry and urinalysis at weeks 5 and 13, gross necropsy evaluations, sperm morphology, and histopathology at study termination.
Results	
Remarks	Male rats treated with methyl propene derivative for 13 weeks at dose levels 250 mg/kg/day gained less weight (15% less at study termination) than controls. Female weights were unaffected. At doses 250 mg/kg/day, both sexes had decreased levels of red blood cells and increased levels of neutrophils in circulation, increased spleen size and increased pigment and red pulp in the spleen. At doses 100 mg/kg/day, there was increased production of WBC in spleen and bone marrow. Mean liver to body weights were increased in male rats at dose levels 250 mg/kg and in female rata at 500 mg/kg/day. Male rats treated with undiluted test material at 500 or 2000 mg/kg/day had increased kidney weights correlated with dose-related increase in hyaline droplet formation indicative of light hydrocarbon nephropathy. Undiluted test material and dilutions at 25% (500 mg/kg, 250 mg/kg in Part 2) induced moderate to strong reaction in the skin, characterized by erythema, edema, increased thickness and stiffness; these effects were more severe in the 500 mg/kg (diluted50%w/v) Microscopically, hyperkeratosis, hyperplasia of sebaceous gland, increased mitosis in epidermis and dermal abscesses were observed. Virtually no irritation was observed in the vehicle control group or in dose groups of 100, 50, 10 mg/kg/day where dilutions were made at 10% or 5 %w/v. No effects on sperm motility or morphology were observed in rats treated with 2000 mg/kg/day. The relative weight increases in livers of higher dose animals of both sexes had no microscopic correlate and is considered an adaptive response to treatment. The increase in kidney weight and hyaline droplet formation in male rats is indicative of light hydrocarbon nephropathy, a condition considered by EPA to be specific to male rats and not predictive of comparable toxicity in humans. [a] Although many changes in hematology parameters can be associated with infections which can occur with severe skin irritation, increased dose related neutrophil production was observed in animals with minimal skin irritation and can be considered a direct effect of the test material.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Conclusions	NOEL for systemic toxicity was 50 mg/kg/day dermal exposure for 13 weeks. The minimally irritating concentration of methyl propene derivative diluted in 100" mineral oil base stock is 10% (100, 50, 10 mg/kg/day)
Data Quality	Reliable without restrictions: Guideline based study performed in accordance with US GLPs.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information). United States Environmental Protection Agency (EPA) 1991. Alpha 2 microglobulin: association with chemically induced renal toxicity and neoplasia in the male rat. P.85 In Risk Assessment Forum, U.S. Govt. Printing Office, Washington, DC
Other	Undated: 12-29-99

AR 201-12549 b 17

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Test Substance	
CAS #	68511-50-2
Chemical Name	1-propene, 2-methyl-, sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl **Sulfide Category.**
Method	
Method/Guideline followed	None cited
Test Type	21 Day Repeated Dose Dermal Toxicity Study
GLP (Y/N)	**N**
Year (Study Performed)	**1979**
Species	**Rabbit**
Strain	New Zealand White
Route of administration	Dermal to shaved skin of backs and sides
Duration of test	**21 days**
Doses/concentration levels	**40 animals tested: 3 treatment** groups (7M/3F, 8M/2F, 6M/4F), 1 untreated control (5M/5F) Group 1: 140 mg/kg/day undiluted test material Group 2: 560 mg/kg/day undiluted test material **Group 3: 2240 mg/kg/day** undiluted test material
Sex	Male and female
Exposure period	
Frequency of treatment	
Control group and treatment	1 untreated control (5M/5F)
Post exposure observation period	
Statistical methods	None cited.
Remarks field for test conditions	Age of animals at initiation: Not specified following at least 2 weeks **acclimation.** Study was **designed to** evaluate local **and** systemic effects of test material when applied dermally. Methyl propene derivative was applied to the shaved backs and sides (approximately 10% of the body surface) of 3 groups of 10 N.Z. White rabbits 5 days per week for 3 weeks at dose levels of 140, 560 or 2240 mg/kg/day of undiluted test material on the same test schedule. The animals were fitted with plastic collars to inhibit ingestion of the test **material,** which was left uncovered on the skin and not removed prior to the next dose. One untreated shaved control group of IV animals was included in the study. Assessments for local and systemic effects included twice **daily**

	(morning and afternoon) clinical observations, skin irritation scoring 5 days per week, weekly body weights, hematology, serum chemistry and urinalysis at pretest and termination, and gross necropsy evaluations at study termination.
Results	
Remarks	All rabbits survived the duration of the test. Body weight changes were within expected ranges and comparable for all groups. Rabbits in all groups had lethargy, ptosis, G.I. disturbances, nasal and ocular discharges and respiratory distress, all more often in the second and third weeks with no discernible pattern of response. Skin responses included slight to moderate erythema and very slight to slight edema during Week 1 for all treated groups. During Week 2 responses in all treated groups were moderate to severe erythema with additional signs of cracked skin, bleeding and discoloration. Edema was slight at the lowest dose and slight to moderate at the higher doses. During Week 3 all treated groups had severe erythema with cracked and bleeding skin, eschar and discoloration and edema was slight to moderate. No irritation was observed in the control group. Urinalysis values were normal in all groups. Several individual and isolated hematological and serum chemistry values were out of expected range but with no discernible treatment related changes in the mean values for all groups. At necropsy, sporadic occurrences of dark lungs and liver, red and bloated intestines, pale kidney or small or gray spleen were noted with no relationship to treatment. Epithelial hyperplasia of the treated skin was observed in all rabbits with the treated groups exhibiting slightly more severe grades of hyperplasia than the control group. The rabbits were grouped by sex at the start of the study. At necropsy six errors in sexing were discovered which resulted in uneven sex distribution within the groups. Since there were no apparent effect differences between the sexes, the study is not considered to be compromised. With no discernable pattern of response in both test and control groups, observed clinical signs are considered to be related to handling. The occurrence of hyperplasia in all groups suggests a relationship to clipping rather to test material administration. However, ***in-life*** dermal observations revealed severe erythema responses in all treated rabbits and none in the sham treated control group-

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68511-50-2
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Conclusions	A NOAEL was not established in this study. The LOEL for clinical signs and systemic toxicity was 140 mg/kg/day dermal exposure for 3 weeks. No minimally irritating concentration was identified by this study.
Data Quality	Reliable with restrictions. Animal age and organ weight data were not included in the report. The test site was not occluded following test material application.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-28-99

AR 201-12549618

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent with guidelines outline in OECD 401
Test Type	Acute oral toxicity
GLP (Y/N)	Y
Year [Study Performed)	**I987**
Species/Strain	Albino rats of the outbred Sprague-Dawley strain
Sex	Male and female
No. of animals/sex/dose	5
Vehicle	Mineral oil-based material
Route of administration	Oral gavage with a syringe and Nelaton catheter. The animals were fasted overnight before dosing.
Remarks field for test conditions	The sample was administered as supplied at a limit dose of 5.0 mg/kg. Following administration, the animals were allowed food and water for the 15-day observation period. The animals were observed three times on the day of dosing and twice on study day two and daily thereafter. Individual weights were recorded on the day of dosage and on the day of termination (day 15). The animals were euthanized by carbon dioxide at the conclusion of the observation period. Gross autopsies were performed on all animals that died during the observation period and on all survivors after day 15.
Results	LD50 > 5.0 gm/kg
Remarks	All animals survived to termination of the experiment (day 15). Decreased activity (3/5 females), diarrhea (1/5 males), salivation (1/5 males and 1/5 females) and apparent urinary incontinence (3/5 females) were noted following dose administration. All animals appeared normal on study days 5-15. Test material did not cause an adverse effect on mean body weight in either sex.
Conclusions	LD50 > 5.0 gm/kg (males and females)
Data Qnality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549619

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent with EPA Health Effects Guideline OPPTS 870.1300
Test Type	Acute inhalation toxicity
GLP (Y/N)	v
Year (Study Performed)	1988
Species/Strain	Mouse (CD- 1 **strain)** Guinea pig (Hartley strain)
Sex	Male and female
No. of animals/sex/dose	5
Vehicle	Mineral oil-based material, dosed as supplied
Route of administration	Aerosol inhalation
Dose	4.3 mg/L (limit study)
Remarks field for test conditions	Two groups ***of*** five mice/sex and five guinea pigs/sex were exposed for 4 hours to the test material (nominal 5 mg/L) as a liquid droplet aerosol generated by a Laskin nebulizer apparatus delivered into a plexi-glass chamber. Also, control group of mice and guinea pigs was exposed to mineral oil in the same manner as the test-material-exposed group except that the test material was not administrated. The details of **the** whole body exposure are consistent with those described in EPA Health Effects Guideline OPPTS 870.1300. The actual exposure concentration as measured by gravimetric analysis was 4.3 mg/L. Particle size analyses were performed once/hour from the **test** material chamber using a cascade impactor. Animal observations for toxicological signs and mortality were recorded every 15 minutes during the exposure, and twice daily for the 14-day observation period. Individual weights were recorded on the day prior to exposure and on days 2,3,5,8 and 14, At **the conclusion** of **the observation period, the** surviving animals were euthanized by exsanguination under general anesthesia. All animals were subjected to gross necropsy (nasal passages, trachea, external surface, all orifices, the cranial cavity, the brain and spinal cord, and all viscera).
Results	LC50 (mice) > 4.3 mg/L; LC50 (guinea pigs) > 4.3 mg/L
Remarks	The mass median aerodynamic diameter for the studies was 1.6 microns with a geometric standard deviation of 2.1 (estimated percent of particles < 10 microns = 100%) One female guinea pig was

	euthanized on study day 7 because of a broken leg, an effect thought to be unrelated to the administration to the test material. All other animals survived the duration of the study. Observations noted during the test material exposure included reduced activity and matted coat. Signs exhibited by the test animals upon removal from the chamber and during the two-hour post-exposure observation period on day 1 included matted coat, yellow fur, yellow ano-gentital staining and nasal discharge. The control groups were generally unremarkable during the exposure and immediately thereafter. During week 1, the test mice exhibited few signs other than matted coat. The test guinea pigs exhibited matted coat and ano-genital staining. During week 2, ano-genital staining was the only remarkable sign in the guinea pigs. No significant difference was noted between the test and control group weights for either species. No gross lesions that could be attributable to the test material were observed in any of the mice or guinea pigs.
Conclusions	Ten of ten CD-1 mice and ten of ten Hartley guinea pigs received a single four-hour whole-body exposure to 4.3 mg/L test material as a respirable aerosol. All animals survived the exposure and the 14-day post-exposure observation period with the exception of a single guinea pig that was euthanized for a broken limb. Signs of treatment included reduced activity and matted coat during the exposure. The treated animals were generally comparable to air-only control animals during the observation period. Body weight values and gross post mortem observations were generally unremarkable for differences between control and treated animals of either species.
Data Quality	Reliable without restriction Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549b20

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical **is also** referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For **more** information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" **in** HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent **with EPA** Health Effects Guideline OPPTS 870.1300
Test Type	Acute inhalation toxicity
GLP (Y/N)	Y
Year (Study Performed)	1988
Species/Strain	**Mouse** (CD-l **Cobs** Swiss **Albino)** Rat **(Sprague-Dawley CD)** Guinea **pig (Hartley)**
Sex	**Male** and **female**
No. of **animals/sex/dose**	5 animals/sex/dose
Vehicle	Mineral oil-based material, dosed as supplied
Route of administration	Aerosol inhalation
Dose	4.3 mg/L (Limit study)
Remarks field for test conditions	Group of five mice/sex, five rats/sex and five guinea pigs/sex were exposed for 4 hours to the test material as a liquid droplet aerosol generated by a Laskin nebulizer apparatus delivered into a plexi-glass chamber. Also, control groups of mice, rats **and guinea pigs were** exposed to mineral oil in the same manner as the test-material-exposed group except that the test material was not administrated. The details of the whole body exposure are consistent with those described in EPA Health Effects Guideline OPPTS 870.1300. The actual exposure concentration as measured by gravimetric analysis was 4.3 mg/L. Particle size analyses were performed once/hour from the test material chamber using **a** cascade impactor. Animal observations for toxicological signs and mortality were recorded every 15 minutes during the **exposure, and** twice daily for the 14-day observation period. **Individual** weights were **recorded** on the day prior to **exposure and on days** 2,3,5,8 and 14. At the conclusion of the observation period, the surviving animals were euthanized by exsanguination under general anesthesia. All **animals** were subjected to gross necropsy **(nasal passages, trachea, external surface, all orifices, the cranial cavity, the brain and** spinal cord, and all viscera).

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: ACUTE TOXICITY

Results	LC50 (mice) > 4.3 mg/L; LC50 (rat) < 4.3 mg/L; LC50 (guinea pigs) > 4.3 mg/L
Remarks	The test animals received an average analytical exposure concentration of 4.3 mg/L test material with a nominal exposure concentration of 100 mg/L. Particle size distribution measurements showed an average mass median aerodynamic diameter of 3.8 microns with an average geometric standard deviation of 1.9 microns. Approximately 93 percent of the aerosol was 10 microns or less in size. Mortality and physical observations: Three female rats dies within a day after exposure. A single male mouse and a single male guinea pig also died on test days 7 and 9, respectively. All other animals survived the duration of the study. Observations noted during exposure included nasal discharge, salivation, closed eyes and wet fur. Signs exhibited by the rats upon removal from the camber and during the two-hour post-exposure observation period on day included numerous secretory responses, labored breathing, rales and wet fur. Also, several of the females showed tremors. One of the female rats dies two hours after exposure. The mice and the guinea pigs were generally unremarkable except for contaminated fur. Two additional rats were found dead the morning after exposure. The surviving rats (both sexes) continued to show responses without a complete recovery during the 14-day post-exposure observation period, including nasal discharge, labored breathing, rales, and contaminated fur leading to alopecia. Body weight: Significant body weight losses were observed following exposure among all three species. The surviving mice and rats began to recover weight within a week after exposure. However, guinea pigs continued to lose weight throughout the first week and did not show a weight gain until the end of the second week. Gross post mortem observations: Discoloration of the lungs and nasal turbinates was noted among the spontaneously dying animals.
Conclusion	LC50 (mice) > 4.3 mg/L; LC50 (rat) < 4.3 mg/L; LC50 (guinea pigs) > 4.3 mg/L
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549 b21

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 403
Test Type	Acute inhalation toxicity
GLP (Y/N)	Y
Year (Study Performed)	1987
Species/Strain	Albino rats of the Sprague-Dawley strain
Sex	Male and female
No. of animals/sex/dose	5 rats/sex/dose
Vehicle	Mineral oil-based material dosed undiluted
Route of administration	Aerosol inhalation
Dose	1.5,2.5 and 5.6 mg/L (actual concentration)
Remarks field for test conditions	Three groups of five rats/sex were exposed for 4 hours to the test material as a liquid droplet aerosol generated by a pressure spray apparatus delivered into a plexi-glass chamber. The details of the whole body exposure are consistent with those described in OECD guideline 403. The actual exposure concentrations as measured by gravimetric analysis were 1.5, 2.5 and 5.6 mg/L. Particle size analyses were performed twice/hour using a multi-stage cascade impactor. Animal observations for toxicological signs and mortality were' recorded periodically during the exposure, and twice daily for the 14-day observation period. Individual weights were recorded on the day prior to exposure and on days 4, 8 and 14. At the conclusion of the observation period, the surviving animals were euthanized using pentobarbital as an anesthetic followed by exsanguination. All animals were subjected to gross necropsy (external body surface and orifices, major visceral organs, body cavities and carcass). The LC50 with 95% confidence intervals was computed using the method of Miller and Tainter (1944).
Results	LC50 (males) > 5.0 mg/L; LC50 (females) = 2.17 mg/L
Remarks	The mass median aerodynamic diameter for the studies was 3.15 microns with a geometric standard deviation of 2.45 (estimated percent of particles < 12 microns = 90.5%). Remarkable animal observations during the studies include alopecia (noted at all dose levels during second week of observation), ataxia (noted prior to the death of one female in the 5.6 mg/L group), dark material around eye

	(noted in two animal/sex at the 5.6 mg/L dose), decreased activity (noted in all animals at the dose level of 2.5 and 5.6 mg/L; reversible by study day 5), respiratory irregularity (increased respiration noted in all groups during and immediately following exposure; reversible by study day 7), tremors (noted in one female during and immediately following exposure to 2.5 mg/L). No male deaths were recorded for any of the dose levels. Group mean body weights were decreased at day 4 among males exposed to 2.5 and 5.6 mg/L. This effect was reversible by study observation day 8 and 14. Three of 5 females in the 1.5 mg/L group died on day 2 following exposure. Four of 5 female rats exposed to 2.5 mg/L died on observation day 2. Three females in the high dose group died on day 2 following exposure, with an addition death on day 6. Body weights decreased at day 4 in the surviving females, an effect that was reversible by days 8 and 14. No internal lesions or abnormalities were noted in any animal sacrificed at study termination. Pathological findings among females which died during the course of the observation period include brain (prominent vascularization, and blood in the cranial cavity), nasal passages (reddening of the nasal passage, with the notation of clear fluid in the nasal passage), lungs (reddening of the lungs, with the observation of a 'puffy' lung in one female) and trachea (clear fluid noted in the trachea of one female).
Conclusion	Following 4-hour whole-body exposure to a liquid droplet aerosol of the test material, the LC50 in male Sprague-Dawley rats is considered to be greater than 5.6 mg/L. The LC50 value in females was calculated to be 2.17 mg/L with upper and lower confidence limits of 3.69 and 0.64 mg/L.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-29-99

AR 201-12549b22

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	FHSA Regulations 16 CFR 1500.40
Test Type	Acute dermal toxicity
GLP (Y/N)	Y
Year (Study Performed)	1986
Species/Strain	Young, adult New Zealand white rabbits
Sex	Male and female
No. of animals/sex/dose	5
Vehicle	Mineral oil-based material dosed undiluted
Route of administration	Dermal application
Remarks field for test conditions	The sample was applied to unabraded shaved skin under impervious occlusion for 24 hours at a limit dose of 2.0 mg/kg At the end of the 24-hour exposure period, the wrapping was removed any unabsorbed test material remaining on the skin was removed by gentle sponging using a paper towel moistened with mineral oil. The animals were observed for signs of toxicity or behavioral changes frequently on the day of treatment. Thereafter, all surviving rabbits were examined for outward signs of toxicity one per day, for the entire 14-day observation period. Individual weights were recorded on the day of dosage, weekly thereafter and prior to sacrifice. The surviving animals were euthanized at the conclusion of the observation period. Gross autopsies were performed on all animals after 14 days.
Results	LD50 > 2.0 gm/kg
Remarks	One rabbit died on day 14 of the study. No other deaths were observed during the 14-day observation period. In the male group, mild skin erythema and mild-t&moderate edema were observed after unwrapping at 24 hours. Slight to mild skin irritation noted at 7 day was completely resolved by day 14. A loss of body weight was noted for 1/5 male animals at day 7. The same animal was found dead on day 14 after experiencing a bloated appearance. Signs of dehydration and no formed fecal material in the intestinal tract were noted for the one mortality. Other than the previous observation, all animals appeared normal throughout the 14-day observation period. In the females, skin reactions were typical of those observed with the male group. A loss of body weight was noted in one female at day 7. Gross pathological

	examination of the female rabbits revealed no remarkable findings.
Conclusions	The test article, when dosed as supplied and studied in 5 males and 5 female albino rabbits, appears to have an acute dermal LD50 greater than 2.0 gm/kg.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549623

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VITRO

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent with guidelines outlined in OECD 471
Test Type	Reverse mutation assay
System of testing	Bacterial
GLP (Y/N)	Y
Year (Study Performed)	1982
Species/Strain	Salmonella typhimurium TA98, TA100, TA1535, TA1537, TA1538
Metabolic activation	With and without
Concentrations	0, 0.01, 0.03, 0.1, 0.3, and 1 microliter/plate (DMSO vehicle)
Statistical methods	The mean number of his- revertants/plate for three replicate assay plates was calculated for each concentration and strain.
Remarks field for test conditions	No significant deviations from guideline protocols
Results	
Remarks	The test material was tested without metabolic activation at 1, 0.3, 0.1, 0.03, and 0.01 microliters of test material/plate and found to be non-mutagenic to the bacterial strains tested. The number of revertant colonies as a result of treatment with the test material did not differ significantly from the number produced by the DMSO vehicle control. The test material was not toxic to any strain at any concentration. The positive controls, sodium azide, 2-nitrofluorene, and 9-aminoacridine at concentrations ranging from 2.5-100 microgram/plate produced more than a lo-fold greater incidence of his' revertants/plate with the bacterial strains used in this study. The test material was also tested in the presence of an S9 microsomal fraction from Aroclor 1254-treated rat livers. The concentrations tested (1, 0.3, 0.l, 0.03, and 0.01 microliters of test material/plate) in the activated system did not induce significant detectable mutagenic events with the bacterial strains used. The positive metabolic activated control, 2-anthramine (2.5 microgram/plate) produced positive mutagenic responses in the bacterial strains used in this study.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VITRO

Conclusions	The test material did not produce significant mutation in any of the Salmonella strains in the quantitative mutagenesis assay, either in the presence or absence of metabolic activation. Thus, under the conditions of the assay employed, the test material was determined to be non- mutagenic in the Salmonella/microsome mutagenesis assay.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549 624

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VIVO

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information **on** the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 474
Test Type	Mammalian erythrocyte micronucleus test
GLP (Y/N)	Y
Year (Study Performed)	1988
Species	Mouse
Strain	B6C3F1
Sex	Male and female
Route of administration	Oral gavage
Doses/concentrations	5 gm/kg (limit dose)
Exposure Period	One dose, dose groups sacrificed after 18,24 and 48 hours
Statistical methods	Group mean body **weights,** total polychromatic erythrocytes (PCEs), normochromatic erythrocytes (NMEs), PCEs with micronuclei, and NMEs with micronuclei were compared. For each animal, a minimum of 1000, PCEs were counted fore the presence of micronucleated PCEs. The frequency of micronucleated cells per animals was expressed as the number of micronucleated PCEs per 1000 PCEs counted The ration of PCEs/NMEs was also recorded. The data were analyzed for statistical significance on a binomial distribution, at a level of significance of 0.05, and using the table of Kastenbaum and Bowman (Mutation Res. 9:527-549, 1970).
Remarks field for test conditions	# of animals per dose: S/sex/group Control groups and treatment: 5/sex negative control (mineral oil); S/sex positive control (cyclophosphamide, 50 mg/kg intraperitoneal injection) Mice were approximately 12 weeks old and 17-31 grams at study initiation. Animals were observed daily and body weights were recorded after 18, 24 and 48 hours. Test material and negative control groups were sacrificed after 18, 24 and 48 hours, whereas the positive control group was terminated after 24 hours.
Results	
Remarks	The frequency of PCEs with micronuclei ranged from 1.0 **to 5.9/1000**

	PCEs in negative control mice with groups means of 2.6, 3.0 and 2.4 PCEs for the three time points. These averages and group means were within were within the expected range based on published data on the performing laboratory historical controls. In contrast, male animals dosed with cyclophosphamide had 9.0 to 24.0 micronucleated PCEs/1000 PCEs, with a mean of 14.5 for the group. The average frequencies of micronucleated PCEs obtained from male animals receiving the test material after the three time periods were 5.1,3.0 and 5.7/1000 PCEs. These group means were not significantly higher than the negative control values. The mean PCE/NME ratios in negative male group for the three time periods were 0.60,0.60 and 0.69, respectively. The test material was not cytotoxic since the PCE/NME ratio at the three time points was 0.60, 0.59 and 0.66. The mean frequency of micronucleated PCEs/1000 PCEs for female mice was 1.9,2.1 and 2.9, respectively. The average micronucleated PCEs value for the cyclophosphamide treated females was 20.5. Female mice treated with the test material were found to have mean micronucleated PCEs values of 1.1,2.0 and 1.2 at the three time points, respectively. A comparison of the PCE/NME ratio between the negative control and test material treated female mice did not vary significantly.
Conclusions	The subject material was tested for its genotoxicity using mouse in vivo micronucleus screening assay in bone marrow. There was no significant increase in micronucleated PCEs in animals exposed to the test substance. Thus, the test material was negative in this assay.
Data Quality	Reliable without restrictions (Klimisch code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-28-99

AR201-12549b25

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 412
Test Type	4-week inhalation toxicity study in rats
GLP (Y/N)	Y
Year (Study Performed)	**1989**
Species	**Rat**
Strain	Sprague-Dawley CD, 7 weeks old at initiation of treatment
Route of administration	Aerosol inhalation
Duration of test	4 weeks of treatment for all doses, and a 3 week recovery period in the control and high dose satellite recovery groups
Doses/concentration levels	0, 15, 50 and 150 mg/m^3
Sex	Males and females
Exposure period	4 weeks of inhalation treatments followed by a 3 week recovery period
Frequency of treatment	Inhalation treatment for 6 hours/day, 5 days/week for 4 weeks at the target concentrations
Control group and treatment	10 rats/sex/group for the low and mid dose levels, 15 male and 20 female rats for the high dose level group. Control rats (15 males and 20 females) received mineral oil only at a level of 150 mg/m^3, while in the exposure chamber.
Post exposure observation period	
Statistical methods	Body weight, food consumption, hematology and clinical chemistry parameters, organ weights and organ/body weight ratios were analyzed. Mean values of ail dose groups were compared to control at each time interval. Tests included parametric ANOVA with a Dunnett's *post-hoc* test, non-parametric Kruskal-Wallis and Dunn's rank sum test, Bartlett's test for equal variances, and Student's t-test.
Remarks field for test conditions	The rats were exposed on each treatment day for 6 hours to the test material (target concentrations = 15, 50, 150 mg/m3) as a liquid droplet aerosol generated by an air atomizing nozzle apparatus delivered into a plexi-glass chamber. Control rats were exposed to in the same manner as the test-material-exposed group except that mineral oil only was administered. The details of the whole body exposure are consistent with those described in OECD guideline 412. The actual exposure concentrations as measured by gravimetric

	analysis were 15, 50 and 160 mg/m^3. Particle size analyses were performed once/week from the test material chamber using a cascade impactor. Animal observations for toxicological signs and mortality were recorded twice daily, once in the morning and once in the afternoon. Over the course of the study. Individual weights were recorded twice pre-test and then weekly during the exposure and recovery periods, and at termination. At the conclusion of the observation period, the surviving animals were euthanized with carbon dioxide. Animals were fasted prior to sacrifice. Five rats/sex were subjected to post-exposure blood analysis (routine hematology and clinical chemistry parameters) on test day 1 for the control and high dose groups, at termination on 5 rats/sex for all dose groups, and on 5 rats/sex from the control and high dose group after three weeks of recovery. Complete gross post mortem examinations were performed on all animals (nasal passages, trachea, external surface, all orifices, the cranial cavity, the brain and spinal cord, and all viscera). Nine major organs were weighed to obtain organ/body weight calculations, 42 individual organs and/or tissues were preserved, and 10 major organs and/or tissues were examined for histopathology.
Results	
Remarks	No NOAEL was assigned to this study. The mass median aerodynamic diameter for the studies ranged from 1.9 to 2.6 microns with a geometric standard deviation ranging from 1.8 to 2.2. This data indicated that the aerosol was of a respirable size in the rat, with at least 96% of the particles 10 microns or less in diameter. Mortality: One high-dose female had convulsive behavior following the third day of exposure, and was found dead the next morning. The cause of death was unclear. There were no other unscheduled deaths in the study. Physical observations: The animals were unremarkable during the exposure period. Weekly detailed observations included an increased incidence of nasal discharge or dried red material on the facial area among the high-dose animals. However, these findings were not temporally consistent nor were they apparent in the lowest two doses of test material. No significant respiratory sounds were noted. Body weights: Although there were no significant differences seen between control and treated groups, there was a trend toward lower body weight gains during the exposure period of the study at all dose levels in the males and with the two highest dose levels in the females. During the three-week recovery period, the high dose animals did not regain the difference in body weight compared to the controls. Hematology: The only significant difference from control values was increased hemoglobin concentration in the high-dose females sacrificed after 4 weeks of exposure. Clinical chemistry: There were several statistically significant differences from the control values at both the post-exposure and post-recovery time intervals. However, these differences

	did not correlate with dose, with sex, with potential target organs or with sacrifice interval. Terminal organ and body weights: Following 4 weeks of exposure to test material, increases in kidney weights were seen in the males at all three dose levels, and were statistically significant in the higher two levels. This effect was considered to renal effects seen microscopically in males (see below). This difference in weight abated following 3 weeks of recovery. Following 4 weeks of exposure, statistically significant increases were seen in high-dose liver weights and liver/body ratio in both sexes. These differences abated following 3 weeks of recovery. Spleen and adrenal weights increased compared to controls in the high dose groups of both sexes. Post-recovery increases in teste, heart, lung and spleen weights were recorded. These effects were not accompanied by pathologic microscopic findings, and therefore, the biological significance was considered equivocal. A few visible gross changes, such as discolored lungs, were noticed in the sacrificed animals. Microscopically, treatment-related effects were seen in the kidneys in the males in a dose-related profile. Findings included globular casts at the cortico-medulary junction, the cortex and medulla, as well as hyaline droplets in the proximal convoluted tubule cells. These responses were seen in males in all treatment groups following 4 weeks of exposure, and in the high-dose group after 3 weeks of recovery. All other microscopic tissue alterations observed in other organs were considered incidental findings.
Conclusions	No NOAEL was assigned to this study.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-28-99

AR 201-12549b26

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 68515-88-8
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Test Substance	
CAS #	CAS# 68515-88-8
Chemical Name	Pentene, 2,4,4-trimethyl-, sulfurized
Remarks	97% purity This chemical is also referred to as trimethyl pentene derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent with OECD 410 and OPPTS 870.3200
Test Type	28-day dermal toxicity study in rats
GLP (Y/N)	Y
Year (Study Performed)	1988
Species	Rat
Strain	Sprague-Dawley CD, 21 days old at initiation of treatment
Route of administration	Test material applied topically to shaved, unabraded skin (semi-occluded dressing)
Duration of test	28 days
Doses/concentration levels	1000 mg/kg/day (limit study)
sex	Males
Exposure period	6 hours/day, after which the test material was removed with mineral oil
Frequency of treatment	5 days/week 4 weeks (total of 20 applications)
Control group and treatment	5 male rats received topical application of test material, 5 male rats served as controls by receiving topical application of mineral oil.
Post exposure observation period	
Statistical methods	Continuous data including body weight, body weight gain and food consumption was analyzed by analysis of variance.
Remarks field for test conditions	
Results	
Remarks	No NOAEL was assigned to this study. All animals survived throughout the study and physical examinations were generally unremarkable. No difference between the test material-treated animals and control animals was noted for the parameters of body weight, body weight gain or food consumption. Detail gross pathological examination of external and internal features of the animals revealed no remarkable findings with the exception of weak-moderate irritation responses at the site of test material application. These responses were characterized by erythema, eschar and flaking of

	the skin that persisted over the majority of the 28-day treatment period.
Conclusions	No NOAEL was assigned to this study. All animals survived throughout the study and physical examinations were generally unremarkable.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-28-99

AR 201-12549 b27

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 67124-09-8
Chemical Name	2-propanol, 1-(tert-dodecylthio)-
Remarks	100% purity This chemical is also referred to as propanol/dodecylthio derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent with guidelines outline in OECD 401
Test Type	Acute oral toxicity
GLP (Y/N)	**Y**
Year (Study Performed)	1982
Species/Strain	Albino rats of the outbred Sprague-Dawley strain
Sex	Male and female
No. of animals/sex/dose	5
Vehicle	Mineral oil-based material dosed undiluted
Route of administration	Oral gavage with a syringe and dosing needle. The animals were fasted overnight before dosing.
Remarks field for test conditions	The sample was administered as supplied at a limit dose of 5.0 mg/kg. Following administration, the animals were allowed food and water for the 14-day observation period. The animals were observed frequently on the day of dosing and twice per day thereafter. Individual weights were recorded on the day of dosage, weekly thereafter and prior to sacrifice. The animals were euthanized by carbon dioxide at the conclusion of the observation period. Gross autopsies were performed on all animals that died during the observation period and on all survivors after 14 days.
Results	LD50 > 5.0 gm/kg
Remarks	The animals were ruffled after 3 hours. They appeared oily and dirty after 24 hours. One death occurred within 48 hours and the remaining animals exhibited a discharge around the eyes and nose. The remaining animals appeared to be recovered by 72 hours. They continued to appear normal throughout the remainder of the observation period. Gross pathological examination reveals no remarkable findings.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
HEALTH ELEMENTS: ACUTE TOXICITY

Conclusions	LD50 > 5.0 gm/kg (males and females)
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549 b28

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
HEALTH ELEMENTS: ACUTE TOXICITY

Test Substance	
CAS #	CAS# 67124-09-8
Chemical Name	2-propanol, 1-(tert-dodecylthio)-
Remarks	100% purity This chemical is also referred to as propanol/dodecylthio derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	FHSA Regulations 16 CFR 1500.40
Test Type	Acute dermal toxicity
GLP (Y/N)	Y
Year (Study Performed)	1991
Species/Strain	Young, adult New Zealand white rabbits
Sex	Male and female
No. of animals/sex/dose	5
Vehicle	Mineral oil-based material dosed undiluted
Route of administration	Dermal application
Remarks field for test conditions	The sample was applied to unabraded shaved skin under impervious occlusion for 24 **hours** at a limit dose of 2.0 mg/kg. At the end of the 24-hour exposure period, the wrapping was removed any unabsorbed test material remaining on the skin was removed by gentle sponging using a paper towel moistened with mineral oil. The animals were observed for signs of toxicity or behavioral changes frequently on the day of treatment. Thereafter, all surviving rabbits were examined for outward signs of toxicity one per day, for the entire 14-day observation period. Individual weights were recorded on the day of dosage, weekly thereafter and prior to sacrifice. The surviving animals were euthanized at the conclusion of the observation period. Gross autopsies were performed on all animals after 14 days.
Results	LD50 > 2.0 gm/kg
Remarks	No deaths were observed during the 14-day observation period. Nasal discharge and fecal staining was observed in 3 of 10 animals. In one animal, the test material cause blistering and blanching at the site of dermal application. Gross pathological examination reveals no remarkable finding.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
HEALTH ELEMENTS: ACUTE TOXICITY

Conclusions	LD50 > 2.0 gm/kg (males and females)
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-21-99

AR 201-12549 b29

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VITRO

Test Substance	
CAS #	CAS# 67124-09-8
Chemical Name	2-propanol, l-(tert-dodecylthio)-
Remarks	100% purity This chemical is also referred to as propanol/dodecylthio derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent with guidelines outlined in OECD 471 and 472
Test Type	Reverse mutation assay
System of testing	Bacterial
GLP (Y/N)	Y
Year (Study Performed)	1988
Species/Strain	Salmonella typhimurium TA98, TA100, TA1535, TA1537 Escherichia coli WP-2
Metabolic activation	With and without
Concentrations	0, 15, 50, 150, 500, 1500 and 5000 microgram/plate (DMSO vehicle)
Statistical methods	Revertant colonies were scored using an electronic colony counter. The mean number of revertants/plate and the standard deviation was calculated for each concentration and swain. A significant effect was considered to be a two-fold increase in revertants when the background was 50 revertant/plate or greater a three-fold increase when the background was between IO and 49 revertants/plate; and a four-fold increase when the background was less than 10 revertants/plate.
Remarks field for test conditions	No significant deviations from guideline protocols
Results	
Remarks	The test material was tested without metabolic activation at 5000, 1500, 500, 150, 50 and 15 microgram/plate and found to be non-mutagenic to the bacterial strains tested. The test material was toxic to TA1537 at 5000, 1500, 500 and 150 microgram/plate. In the confirming assay, the ***test*** material was tested at the identical concentrations, and again, no mutagenic response was observed with any of the bacterial strains. The positive controls, sodium azide, 2-nitrofluorene, 9-aminoacridine, and ENNG at concentrations ranging from 1.0-80 microgram/plate, produced statistically significant positive responses in the bacterial strains used in this study. The test material was also tested in the presence of an S9 microsomal fraction from Aroclor 1254-treated rat livers. The concentrations tested (5000, 1500, 500, 150, 50 and 15 microgram/plate) in the activated system did not induce detectable mutagenic events with the bacterial strains used. The negative responses were reproduced a second confirmatory assay.

	Incidentally, the S9 mix reduced the toxicity of the test material in the presence of TA1537. The positive metabolic activated control, 2-anthramine at concentrations ranging from 0.5-20 microgram/plate, produced statistically significant positive mutagenic responses in the bacterial strains used in this study.
Conclusions	The test material was assayed for its ability to induce mutations in Salmonella typhimurium and Escherichia coli in the presence and absence of a metabolic activation system. At the concentrations tested and under the conditions of the assay, the test material was considered to be non-mutagenic.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549 630

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VITRO

Test Substance	
CAS #	CAS# 67124-09-8
Chemical Name	2-propanol, 1-(tert-dodecylthio)-
Remarks	100% purity This chemical is also referred to as propanol/dodecylthio derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	Consistent with EPA guidelines outlined in OPPTS 870.5375
Test Type	In vitro chromosomal aberration assay
System of testing	Non-bacterial
GLP (Y/N)	**Yes**
Year (Study Performed)	1989
Species/St&n	Chinese hamster ovary (CHO) cells
Metabolic activation	S9 fraction prepared from livers of Aroclor 1254-induced Sprague-Dawley rats
Concentrations	Non-activated assay: 0, 0.05, 0.15, 0.5, 1.5, 5, 15, 50, 495, 1490, 4950 **ug/ml** Activated assay: 0, 0.05, 0.15, 0.5, 1.5, 15, 50 and 150 ug/ml
Statistical methods	Metaphase cells were analyzed for chromosomal aberrations by 100x objective microscopy. The mitotic index was determined by counting a minimum of 500 total cells. Coordinates of cells with aberrations were recorded. The data were analyzed statistically using the method described in Margolin et al., Statistical analysis for in vitro cytogenetic assay using Chinese hamster ovary cells, Environ Mutagen 8:183-204, 1986.
Remarks field for test conditions	No significant deviations from guideline protocols
Results	
Remarks	The test material was investigated for its ability to induce chromosomal aberrations in Chinese hamster ovary (CHO) cells in the presence and absence of a rat liver homogenate metabolic activation system. CHO cells were seeded at a density of OS or 0.75 x 10^6 and maintained in essential culture medium in tissue culture flasks. Twenty-four hours later, the cells were exposed to test material, positive or negative controls. The compound was dissolved in DMSO, which served also as the negative control. The test concentrations that were tested for the induction of aberrations ranged from 0.05 to 4950 ug/ml in the non-activated assays, and from 0.05 to 150 ug/ml in the activated assays. All test sample concentration and controls were tested in duplicate flasks. Two assay periods were used, 10 and 20 hours, which totaled four independent experiments. The data were analyzed using two methods. Two hours prior to harvest, vinblastine

	sulfate was added to arrest the cells in metaphase. At the end of the incubation period, metaphase cells were collected by treatment with trypsin, concentrated by centrifugation, lysed in hypotonic solution, fixed in methanol: acetic acid and stained with Giemsa. The first used the traditional method in which gaps were not counted as chromosomal aberrations while the second method counted gaps as chromosomal aberrations. One hundred cell were scored from each duplicate flask for each concentration tested. The test substance was toxic to the CHO cells in the non-activated assays at concentrations higher than 15 ug/ml for the 20-hour exposure period (86% reduction in the mitotic index), and at 50 ug/ml for the lo-hour exposure period (.90% reduction of the mitotic index). There was no significant increase in the percentages of aberrant cells in the 10 (4% vs. 4.5% vehicle control) and 20-hour (4% vs 3% vehicle control) non-activated assays. In contrast, the positive control mitomycin C (0.3 ug/ml) caused a significant increase in the percentage of cells with aberrations (approximately 85%). In the activated assays, cells were exposed simultaneously to test material and S9 microsomal fraction with isocitrate cofactors for 10-20 hours. After this period the cells were washed, re-incubated for 8 hours prior to metaphase arrest and chromosomal staining. Concentrations greater than 50 ug/ml for the 20-hour period and 15 ug/ml for the 10-hour exposure period were toxic (> 90% reduction in mitotic index for each incubation period). There was no increase in the percentage of aberrant cells in the 20-hour activated experiment (4.6% vs 4.5% vehicle control). A slight increase in aberrant cells was observed at 5 ug/ml in the 10-hour activated assay, however, this increase was not statistically significant (17.8% vs 14.5% vehicle control). The positive control (benzo[a]pyrene; 15 ug/ml) caused aberration in nearly 100% of CHO cells in the 20-hour activated assay and 46% in the 10-hour activated assay, thus, demonstrating the efficacy of the metabolic activation system.
Conclusions	The test material was assayed for its ability to induce chromosomal aberrations in in vitro culture of Chinese hamster ovary cells in the presence and absence of a metabolic activation system. At the concentrations tested and under the conditions of the assay, the test material was considered to be non-clastogenic.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

AR 201-12549 b31

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Test Substance	
CAS #	CAS# 67124-09-8
Chemical Name	2-propanol, l-(tert-dodecylthio)-
Remarks	100% purity This chemical is also referred to as propanol/dodecylthio derivative in the HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 407
Test Type	28-day oral toxicity study in rats
GLP (Y/N)	Y
Year (Study Performed)	1991
Species	Rat
Strain	Sprague-Dawley CD, 41 days old at initiation of treatment
Route of administration	Oral gavage (syringe and dosing tube)
Duration of test	28 days of treatment and 14 day recovery period in the control and high dose satellite recovery groups
Doses/concentration levels	0, 100, 300 and 1000 mg/kg/day
Sex	Males and females
Exposure period	28-day treatment duration with a 14 day recovery
Frequency of treatment	7 days/week
Control group and treatment	5 rats/sex/group for each dose, and satellite recovery groups of 5 animals/sex for the control and 1000 mg/kg/day dose. Control group received daily doses of corn oil at 2.0 ml/kg, and treatment groups received the indicated dose of test material diluted in corn oil in a volume not to exceed 2.0 ml/kg
Post exposure observation period	14-days
Statistical methods	Body weight, food consumption, hematology and clinical chemistry parameters, organ weights and organ/body weight ratios were analyzed. Mean values of all dose groups were compared to control at each time interval. Tests included parametric ANOVA with a Dunnett's *post-hoc* test, non-parametric Kruskal-Wallis and Dunn's rank sum test, Bartlett's test for equal variances, and Student's r-test.
Remarks field for test conditions	Significant deviations from the OECD 407 test guidelines include: • A function observational battery for neurotoxicity was not performed since this test was not part of the OECD 407 guideline at the time the study was performed
Results	
Remarks	No NOAEL was assigned to this study. All animals survived throughout the study and physical examinations

were generally unremarkable. Test material administration produced alterations in the liver and kidneys of treated animals that were evident in the evaluation of organ weights as well as gross and microscopic pathological examinations.

Dose-related elevations in mean liver weights and/or liver/body weight ratios were seen at study termination in males at all dose levels and in females at the mid- and high-dose levels. Recovery was apparent during the two-week recovery period for the high-dose group. Gross post mortem examination of the liver revealed an accentuated lobular pattern in the mid- and high-dose females at termination of the dosing period, which resolved during the recovery period. Microscopic examination of liver revealed hepatocyte hypertrophy in all dose groups at the termination of treatment. This effect continued through the recovery period. The effect on the liver was consistent with the adaptive induction of hepatic metabolic mechanisms in response to a xenobiotic challenge.

Kidney alterations were seen only in males. Kidney weights and kidney/body weight ratios for high-dose males were significantly higher than control values at termination of dosing. These values were comparable following termination of the recovery period. Gross post mortem examination of the kidneys revealed pale or tan discoloration of increasing frequency with increased dose. Microscopic alterations consisted of increased incidences of globular casts and hyaline droplets in treated males. Hyaline droplets in the proximal tubules were seen at termination of dosing only, indicating that this change in renal morphology was reversible after cessation of test substance administration. The renal effects are consistent with previous reports in the scientific literature of male rat-specific hydrocarbon nephropathy. Evaluation of clinical chemistry and urinalysis studies revealed no evidence of renal or hepatic functional alterations, or any other signs of systemic effects due to the test material. Other minor effects of the test material consisted of a transient decrease in food consumption and body weight gain in the high-dose male group during the first week of study. A slight decrease in hemoglobin and hematocrit values was observed in the high-dose female group at termination that was found to be reversible during the 2-week recovery period.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
HEALTH ELEMENTS: REPEATED DOSE TOXICITY

Conclusions	Although renal and hepatic changes were evident at all dose levels (100,300, and 1000 mg/kg/day), the renal changes are species-specific and the hepatic changes are probably adaptive in nature. Therefore, little subchronic toxicity was observed over the range of doses administered in this study.
Data Quality	Reliable without restriction (Klimisch Code)
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12-27-99

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS:
BIODEGRADATION

Test Substance	
CAS #	67124-09-8
Chemical Name	2-propanol, 1-(tert-dodecylthio)-
Remarks	This substance is also referred to as propanol/dodecylthio derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 301F
Test Type (aerobic/anaerobic)	Aerobic
GLP (Y/N)	Yes
Year (Study Performed)	1998
Contact time (units)	28 days.
Inoculum	Return activated sludge from domestic waste water treatment plant.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS: BIODEGRADATION

Remarks for test conditions	Inoculum: The sludge was aerated and stirred for 2-3 hours, homogenized for 2 minutes and allowed to stand for one-half to one hour. The supernatant was pipetted out and used for pre-adaptation. The inoculum was pre-adapted to the test material for 14 days during which the test substance was added incrementally at concentrations equivalent to 4, 8 and 8 mg carbon/L. on days 0, 7, and 12, respectively. The targeted microbial level in the test mixture was 10,000 to 1,000,000 cells/ml. Conc of test chemical: Test substance concentration was approximately 100 mg/L mineral medium, giving at least 50 to 100 mg ThOD per L medium. No organic solvents were used to facilitate the dispersion of the test material. The test substance was weighed onto a teflon coupon and introduced into the medium. Temp of incubation: 23 ± 1°C. Dosing procedure: A measured volume of the inoculated mineral medium containing approximately 100 mg/L test substance is continuously stirred in a closed system for 28 days. Sampling: The oxygen uptake were monitored continuously and recorded every 4 hours throughout the test. Controls: Yes, except abiotic and toxicity checks were not included. Analytical method: Oxygen uptake was measured using a BI-1000 electrolytic respirometer system. Method of calculating measured concentrations: Not applicable. Other: The inoculum was pre-adapted to the test substance for 14 days.
Results	
Degradation % after time	5.9% after 28 days.
Kinetic (for sample, positive and negative controls)	% biodegradation (days) Reference (sodium benzoate) - 30.5% (1d), 76.4% (7d), 88.8% (28d). Test substance - 0% (1d), 1.6% (7d), 5.9% (28d).
Breakdown Products (Y/N) If yes describe breakdown products	Not monitored.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS: BIODEGRADATION

Conclusions	The test substance showed a low biodegradation rate (5.9%) in 28 days. The reference substance, sodium benzoate, reached a level of 88.8% in the same test period.
Data Quality	Reliable without restrictions
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12/27/99

ROBUST SUMMARY
ALKYL SULFIDE CATEGORY
CAS #68511-50-2
GENETIC TOXICITY ELEMENTS: GENETIC TOXICITY IN VITRO

Test Substance	
CAS#	68511-50-2
Chemical name	1-propene, 2-methyl- sulfurized
Remarks	This substance is also referred to as methyl propene derivative in HERTG's Test Plan for the Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/guideline followed	Consistent with guidelines outlined in OECD 471
Test Type	Reverse Mutation Assay
System of testing	Bacterial
GLP (Y/N)	No
Year (study performed)	1978
Species/Strain	*Salmonella typhimurium* strains TA1535, TA100, TA1537, TA1538, and TA98
Metabolic activation	Test conducted with and without metabolic activation Adult male Sprague-Dawley rat liver S-9 fraction, induced with Aroclor 1254 100 ul/plate
Concentrations	0, 0.01, 0.05,0.1, 0.5, 1.0 ul of test agent per plate with and without metabolic activation
Statistical Methods	Determination of mean ±S.D. of replicate plate counts
Remarks Field for Test Conditions	The vehicle was DMSO; All stock and working solutions were stored at 4°C in glass screw-capped bottles; All sterility controls were negative for bacterial growth; Vehicle was tested as negative control; Positive controls (9-aminoacridine and 2-nitrofluorene without activation and 9-aminoacridine, 2-nitrofluorene, aflatoxin, and 6-aminochrysene with activation) were at least 3 times the number of colonies as the control.
Results	
Remarks	For all strains and dose levels with and without metabolic activation, the criteria for a positive mutagens (at least 3 times the number of colonies as the controls for spontaneous reversion) was not met.

Conclusions	The test agent did not induce a significant increase in the number of point mutations in *Salmonella typhimurium* strains in the absence of the activating system for strains TA1535, TA100, TA1537, TA1538, and TA98. It also did not induce a significant increase in the number of point mutations with the addition of an exogenous source of liver enzymes for metabolic activation in strains TA1535, TA100, TA1537, TA1538, and TA98.
Data Quality	Reliable. Comparable to guideline study.
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 4-12-00

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS: BIODEGRADATION

Test Substance	
CAS #	67124-09-8
Chemical Name	2-propanol, 1-(tert-dodecylthio)-
Remarks	This substance is also referred to as propanol/dodecylthio derivative in HERTG's Test Plan for Alkyl Sulfide Category. For more information on the chemical, see Section 2.0 "Chemical Description of Alkyl Sulfide Category" in HERTG's Test Plan for Alkyl Sulfide Category.
Method	
Method/Guideline followed	OECD 301F
Test Type (aerobic/anaerobic)	Aerobic
GLP (Y/N)	Yes
Year (Study Performed)	1998
Contact time (units)	28 days.
Inoculum	Return activated sludge from domestic waste water treatment plant.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS: BIODEGRADATION

Remarks for test conditions	Inoculum: The sludge was aerated and stirred for 2-3 hours, homogenized for 2 minutes and allowed to stand for one-half to one hour. The supernatant was pipetted out and used for pre-adaptation. The inoculum was pre-adapted to the test material for 14 days during which the test substance was added incrementally at concentrations equivalent to 4, 8 and 8 mg carbon/L on days 0, 7, and 12, respectively. The targeted microbial level in the test mixture was 10,000 to 1,000,000 cells/mL. Conc of test chemical: Test substance concentration was approximately 100 mg/L mineral medium, giving at least 50 to 100 mg ThOD per L medium. No organic solvents were used to facilitate the dispersion of the test material. The test substance was weighed onto a teflon coupon and introduced into the medium. Temp of incubation: 23 $\pm$ 1ºC. Dosing procedure: A measured volume of the inoculated mineral medium containing approximately 100 mg/L test substance is continuously stirred in a closed system for 28 days. Sampling: The oxygen uptake were monitored continuously and recorded every 4 hours throughout the test. Controls: Yes, except abiotic and toxicity checks were not included. Analytical method: Oxygen uptake was measured using a BI-1000 electrolytic respirometer system. Method of calculating measured concentrations: Not applicable. Other: The inoculum was pre-adapted to the test substance for 14 days.
Results	
Degradation % after time	5.9% after 28 days.
Kinetic (for sample, positive and negative controls)	% biodegradation (days) Reference (sodium benzoate) – 30.5% (1d), 76.4% (7d), 88.8% (28d). Test substance – 0% (1d), 1.6% (7d), 5.9% (28d).
Breakdown Products (Y/N) If yes describe breakdown products	Not monitored.

ROBUST SUMMARY
ALKYL SULFIDE CATETGORY
CAS # 67124-09-8
ENVIRONMENTAL FATE AND PATHWAY ELEMENTS: BIODEGRADATION

Conclusions	The test substance showed a low biodegradation rate (5.9%) in 28 days. The reference substance, sodium benzoate, reached a level of 88.8% in the same test period.
Data Quality	Reliable without restrictions
References	This robust summary was prepared from an unpublished study by an individual member company of the HERTG (the underlying study contains confidential business information).
Other	Updated: 12/27/99

HIGH PRODUCTION VOLUME (HPV) CHALLENGE PROGRAM

Summary of Boiling & Melting Points for Members of ALKYL SULFIDE CATEGORY

CAS #	Value	Method *	Year	Remarks
67762-55-4	186.9 °C melting point	calculated by MPBPWIN (v1.31)	1999	y=2.5, x=1: melting point = 186.92 °C y=2.5, x=2: melting point = 213.83 °C
67762-55-4	> 300 °C boiling point	calculated by MPBPWIN (v1.31)	1999	y=2.5, x=1: boiling point = 504.54 °C y=2.5, x=2: boiling point = 537.80 °C
68511-50-2	147.5 °C melting point	calculated by MPBPWIN (v1.31)	1999	y=3: melting point = 147.52 °C y=8: melting point = 329.32 °C
68511-50-2	> 300 °C boiling point	calculated by MPBPWIN (v1.31)	1999	y=3: boiling point = 409.48 °C y=8: boiling point = 749.88 °C
68515-88-8	128.1 °C melting point	calculated by MPBPWIN (v1.31)	1999	y=1: melting point = 128.08 °C y=4: melting point = 186.87 °C
68515-88-8	> 300 °C boiling point	calculated by MPBPWIN (v1.31)	1999	y=1: boiling point = 377.79 °C y=4: boiling point = 477.56 °C
72162-15-3	58 °C melting point	calculated by MPBPWIN (v1.31)	1999	Dimer S: melting point = 58.01 °C Dimer SS: melting point = 88.67 °C
72162-15-3	> 300 °C boiling point	calculated by MPBPWIN (v1.31)	1999	Dimer S: boiling point = 353.69 °C Dimer SS: boiling point = 386.95 °C
67124-09-8	68.15 °C melting point	calculated by MPBPWIN (v1.31)	1999	
67124-09-8	302.85 °C boiling point	calculated by MPBPWIN (v1.31)	1999	

This robust summary was prepared from modeled data by an HERTG member company representative.
Reliability: (2) valid with restrictions

* Reference: Meylan W. and Howard P. 1999. EPIWin Modeling Program, Syracuse Research Corporation. Environmental Science Center, 6225 Running Ridge Road, North Syracuse, NY 13212-2510